Journal of the
OPTICAL SOCIETY
of AMERICA

VOLUME 34, NUMBER 8 AUGUST, 1944

On Plate Calibration in Spectrographic Analysis*

SAUL LEVY
Carnegie-Illinois Steel Corporation, Gary Works, Gary, Indiana
(Received May 23, 1944)

1. METHODS OF PHOTOGRAPHIC PHOTOMETRY

THERE are, essentially, two procedures in photographic radiation measurements in use. The first is based on the so-called Hartmann principle,[1] namely, that two sources of equal intensity and of the same wave-length produce equal blackenings on the same plate if the time of the exposure is the same. This principle sounds trivial, and yet it is the basis of modern photometric procedure, as applied first by Koch[2] in his measurements of line broadening. Using sources of known intensity ratios, one obtains intensity marks on the plate and by measuring the density (or transparency) of the plate, the whole density-intensity curve is obtained. The second method involves the application of some kind of law for the density of the plate as a function of exposure, such as the linear approximation of the Hurter and Driffield curve (density-intensity relation).

The photometric procedures used in spectrographic analysis are similar to those mentioned above (see, however, Section 4). In one method the intensities of spectral lines which depend on the concentrations of the elements in the sample are measured with the help of an empirical characteristic curve. In the other method use is made of a linear approximation for the "normal" blackening portion of the H & D curve, using standards to account for the changeable contrast.[3]

The first method is in principle correct; however, it is not always convenient in quick routine work, as it requires several measurements and careful plotting of the characteristic curve. Often in practice this is not made for each plate and the curve is only checked from time to time. However, the contrast of the plates might differ occasionally even on plates taken from the same box, because of conditions of storage, developing, etc.

The second method is a rapid one and can be applied for each plate. The linear approximation, however, might not always be sufficient, especially, when a larger density range is used.

In the present paper a simplified method of plate calibration is described which is believed to be satisfactory in many cases occurring in practical work.

2. ANALYTICAL REPRESENTATION OF THE CHARACTERISTIC CURVE

The valuable feature of the second of the above-described methods (using the linear approximation of the H & D curve) is that it permits the calibration of the plate by measuring the densities of one pair of lines of sufficient stability, as the

*Abstract in Phys. Rev. 65, 256 (1944).

[1] J. Hartmann, Zeits. f. Instrumentenk. 19, 97 and 842 (1899).

[2] P. P. Koch, Ann. d. Physik 30, 841 (1909); 34, 377 (1911).

[3] This procedure is described in F. Twyman's *Spectrochemical Analysis of Metals and Alloys* (Chemical Publishing Company, Brooklyn, New York, 1941), p. 97.

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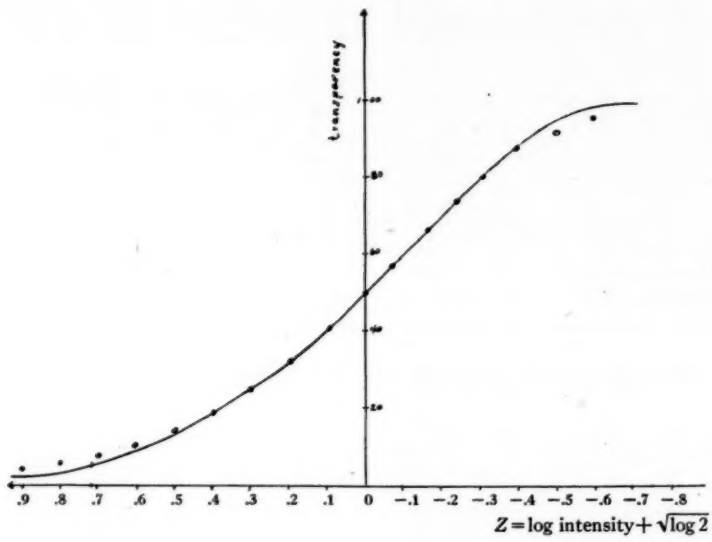


FIG. 1.

measured intensity ratio determines the only constant of the linear equation important for relative measurements, namely, the slope. The question arises whether the actual characteristic curve, in its usable part at least, depends also on only one constant. If that is the case, then the calibration can be obtained by measuring one intensity ratio, this single constant determining the whole characteristic curve of the plate.

It is of no decisive importance for our practical purposes whether or not an explicit analytical expression can be found which represents the calibration curve with a sufficient accuracy. Nevertheless, it is interesting to note that theoretical considerations have led to more or less complicated expressions for the H & D curve. The expression obtained by Silberstein[4] contains only one constant essential for relative measurements. Since the theoretical interpretation of this constant encounters some difficulties, Silberstein considers his expression as a semi-empirical one, as it agrees well with the experimental results with pure silver-bromide emulsions. Webb,[5] in a somewhat different way, arrives at the same conclusion, namely, that the theoretical H & D curve contains only one essential constant.

Perhaps the simplest of all analytical expressions suggested for the representation of the characteristic curve is that given by Abney, which for our purposes can be written: $u/u_0 = \exp[-(k \log I)^2]$, where u/u_0 is the transparency, I—intensity of light, and k—a constant. It is not considered to be a good approximation[6] for the whole extension of the curve; however, for that part of the curve which is mostly used for spectrographic purposes with transparencies between 0.10 and 0.80, or densities 1.0 and 0.1, respectively, the agreement between the calculated and the experimental curves is fairly close, as it was found in this laboratory (Fig. 1) for Eastman Kodak Spectrum Analysis No. 1 plates, using the light of a spectral line of wave-length 3100, from a spark. The circles are the mean values of actual measurements, the curve is one wing of the normal frequency distribution, with $z = \log I$ and k adjusted for the middle portion of the curve. The experimental and the calculated

TABLE I.

Transparency		Intensity ratios	
First line	Second line	Calc.	Experimental
0.50	0.40	0.79	0.79
0.50	0.30	0.60_5	0.60_5
0.50	0.20	0.42	0.42
0.50	0.15	0.34	0.33
0.50	0.10	0.26_5	0.24
0.50	0.05	0.16_5	0.15

[4] L. Silberstein, J. Opt. Soc. Am. **32**, 474 (1942).
[5] J. H. Webb, J. Opt. Soc. Am. **29**, 314 (1939).
[6] C. K. Mees, *The Theory of Photographic Process* (D. Van Nostrand Company, Inc., New York, 1943).

curves are brought to coincidence at their inflection points, where their values of the ordinate is 0.50. Between the transparency 0.85 and 0.20 the agreement is excellent. The deviation starts at about 0.15 and is not negligible.[7] Table I gives the intensity ratios computed from these two curves for different transparencies.

It appears thus that the characteristic curve can be represented very closely by expressions having one constant. By changing this constant which, obviously, is characteristic of the plate contrast, one obtains a family of curves for different contrast. For instance, by variation of k in the Abney approximation one obtains curves which deviate one from another only in the unit of the abscissa, that is, of the log intensity axis. In general, as it will be shown in Section 3, the characteristic curves for plates of different contrasts can be found by changing the unit of the log intensity axis, when the curve for one particular contrast is found experimentally. The applicability of those curves depends on the validity of the following assumptions: (1) The transparency of a photographic plate is always the same monotonous function of the exposure. (2) This function contains only one constant. This implies the assumption that all the factors other than exposure, such as temperature and properties of the developer, time of developing, and so forth, exert a combined influence on one single physical quantity, responsible for the blackening of the plate. In Silberstein's theory it is the minimum number of light quanta which render the grain developable.

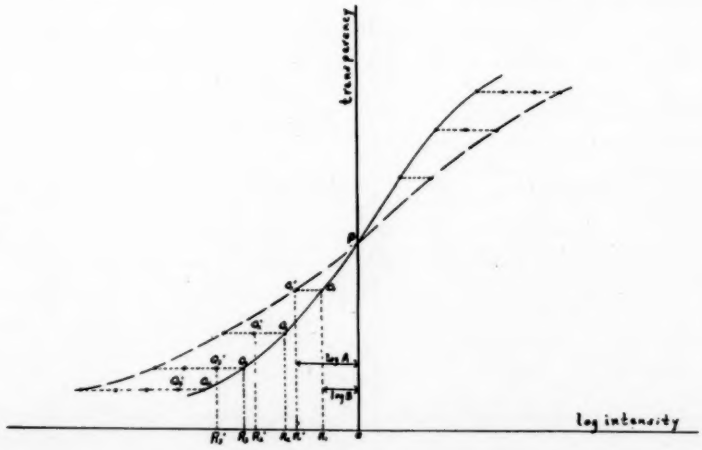
Whether and to what extent the curves thus obtained actually represent the characteristic curves for the plates of different contrasts can be decided by comparison with the experimental curves or by investigating the consistency of the measured intensity ratios on plates of different contrasts, applying those curves.

3. CHARACTERISTIC CURVES FOR PLATES OF DIFFERENT CONTRASTS

Let us suppose that the characteristic curve for one particular plate was found experimentally. By selecting a pair of lines (of the matrix element, for instance, of iron, when steel is to be analyzed) of different brightness and of a stable intensity ratio, one obtains a certain value for the latter, applying the experimental characteristic curve of the plate to the transparencies of the lines. For convenience, we shall call this experimental curve the basic curve, the selected pair of lines—the gamma-pair. The measured intensity ratio of this pair, the gamma-pair ratio of the plate, should have the value A. The application of the basic curve to the blackenings of the gamma-pair lines on other plates may yield ratios different from A, on account of a different characteristic curve corresponding to those plates. Suppose that for a particular plate

[7] It should be emphasized that the measured transparency depends to a certain extent on the type of densitometer used: the difference between the "specular" and the "diffuse" transparency, as a result of the scattering of light in the emulsion. The theoretical notion of transparency (or density), as well as the measured values of this quantity are therefore somewhat indefinite.

FIG. 2.



the gamma-pair ratio, as measured with the basic curve, is B instead of A. A characteristic curve is sought which being applied to the gamma-pair transparencies of that plate yields the value A. With the assumption made in Section 2, the following construction can be made. Let the full curve (Fig. 2) be the basic characteristic curve, PO and Q_1R_1 the transparencies of the gamma-pair lines on that plate, and $OR_1 = \log B$. As the shift of the characteristic curve with log intensity scale does not change the relative intensities, we may assume that the curve to be constructed intersects the basic curve at the point P. The transparencies PO and Q_1R_1 should have abscissa difference $OR_1 = \log A$, when the new curve is applied. Therefore the point Q_1^1 with the ordinate $Q_1R_1 = Q_1^1R_1^1$ and the abscissa $OR_1^1 = \log A$ belongs to the curve in question. We consider now the transparencies Q_1R_1 and Q_2R_2 with the abscissa difference $R_1R_2 = OR_1 = \log B$. Assuming now that the new curve is shifted to the right the distance $R_1^1R_1$, and intersects now the basic curve at Q_1, we find in a similar way the point Q_2^1. Considering the transparencies Q_2R_2 and Q_3R_3 with the same abscissa difference $R_2R_3 = R_1R_2 = OR_1$, and assuming that the new curve is again shifted to the right the distance $R_2^1R_2$, so as to make the intersection point of two curves at Q_2, we obtained the point Q_3^1. In this way we obtain a series of points Q_1^1, Q_2^1, Q_3^1, etc. By shifting to the left the point Q_2^1 the distance $R_1R_1^1 = R_2^2R_2$, Q_3^1—twice this distance, Q_4^1—three times, and so on, we obtain the points of a curve which intersects the basic curve at P and which yields in all its parts the intensity ratio A applied to transparencies of a plate, when the basic curve gives the ratio B.

It will be noticed that the new curve can be obtained from the basic one by changing the abscissa unit in the ratio $\log A / \log B$.

The assumption made in Section 2, namely, that the characteristic curves are determined by measurement of two transparencies, implies that the constructed curve is also applicable to intensity ratios other than that of the gamma-pair. That is, the curve is the characteristic curve for all plates on which the gamma-pair measured with the basic curve has the value B.

The same construction can be made for different values of the gamma-pair ratio as measured with the basic curve. In this way a set of calibration curves is obtained, in the range of expected variations of the contrast and at intervals desired. While determining the relation between the intensity ratios and concentrations and then for concentration determinations, one has to measure, first, the transparencies of the gamma-pair and to find the intensity ratio with the basic calibration. The resulting value indicates which calibration must be used for the plate in question.

As the calibrations are made for permanent use, the two-dimensional diagrams can be transformed into one-dimensional scales by marking the transparencies on the corresponding log

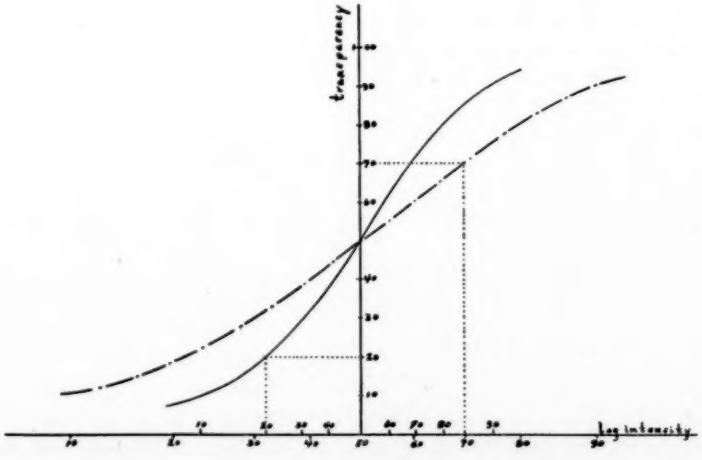


FIG. 3.

intensity axis (Fig. 3), in a similar way, as suggested by Owens[8] for the concentration—log intensity relations (analytical scales) and used in conjunction with them. When the dark and the clear-plate galvanometer deviations are adjustable to 0 and 100, respectively, and the deviations are proportional to the transparencies, they can be used instead of transparencies.

Needless to say, the empirically found basic characteristic curve must be made with all possible care, averaging the results from several plates of the same contrast, as any error in the basic curve will be reflected in all the others. A correctly drawn characteristic curve should give intensity ratios independent of the absolute brightness of the source.

There are several methods of preparing the characteristic curves described in the literature. In the next section a procedure is described which does not require the application of any device such as step filter, with known transparency ratios.

4. PROCEDURE FOR OBTAINING THE BASIC CURVE

The requirements on plate calibration for spectrographic analysis are different from those for general photographic radiation measurements.

Measuring the true intensity ratios, for instance, in multiplets, is a considerable task. The plate calibration is made with some kind of step weakener with accurately known transmission ratios.[9] The preparation of the weakener and the measurement of the transparency for ultraviolet light is not a simple matter, especially in a laboratory not equipped for work of that kind. Furthermore, the relatively long spectrographic slit required for the use of several filter steps must be very uniformly illuminated, which requires special care. For each wave-length a separate characteristic curve is necessary.

A different problem is encountered in spectrographic analysis. The goal is not the intensity ratios; they are only a means for concentration determination. From the relation between intensity and transparency (or density) and the relation between intensity and concentration, the intensity has to be eliminated, which is often made by means of a graphical procedure.[10] Not only the unit of the intensity scale, but also the true values of the intensity ratios (of the impurity line and the matrix line) are of no significance. The intensity ratios can be distorted by a constant factor, the same for the characteristic curve and for the concentration calibration. Only the cross ratios are of significance. Accordingly, not the transparency ratios of the neighboring steps of light weakener (filter, step sector, etc.) but their cross ratios must be known. In the simplest case the cross ratios may be constant: The transparency ratio of the first step to that of the second may be the same, as the ratio of the second to the third, and so forth.

Therefore, when no step filter or a similar device with known transparency cross ratios is available or its use not desirable, the following procedure leads to satisfactory results. Several spectrograms are made on the same plate with the same time of exposure, using different light filters, such as sieves of different mesh numbers, or some other light weakener. Their transparency need not be known. A pair of lines of the matrix element, which can be used as a gamma-pair, of different brightness and of a sufficient stability is selected, and an arbitrary "intensity ratio" is assigned to this pair of lines. An approximate value of the ratio can be found, if desired, by making spectrograms with different exposure and applying the reciprocity law. No use will be made later of this value other than for characterizing the contrast. The blackenings of these two lines can be used as "steps" for the characteristic curve of the plate. If the light filters used happened to be of such a transparency that the transparencies (or the galvanometer deviations of the microphotometer) of the stronger line on one of the spectrograms is equal to the transparencies (or the galvanometer deviations) of the weaker line on another spectrogram, and so forth—an ideal case, of course—then the measurements can be used for plotting the characteristic curve immediately. An illustrative example: Let us suppose, that the galvanometer deviations of these two lines in the consecutive spectrograms were:

Weaker line	Stronger line
90	73
73	51_5
51_5	30_5
30_5	16
16	8_5
8_5	4.

[8] J. S. Owens, Metals and Alloys **9**, 15 (1938).
[9] See, for instance, Dorgelo, Physik. Zeits. **26**, 756 (1925).
[10] J. S. Owens, reference 8.

As the intensity ratio of the lines is assumed to be stable, a curve can be drawn with the galvanometer deviations as ordinates and equidistant abscissa values using an arbitrary unit of length. This corresponds to a characteristic curve with log intensity as abscissa. The curve satisfies the condition that the "intensity ratios" of the measured lines—that is, the distances on the log intensity scale, corresponding to their transparencies, as determined with this curve—are independent of the absolute brightness of the source, by virtue of its construction.

The coincidence of the galvanometer deviations of the weak and the strong lines in the different spectrograms is not likely to occur; it is, however, not necessary because interpolation can be applied. If instead of the readings 51_5 and 30_5 in the above example, 55 and 33 were obtained, then a point corresponding to the ordinate 55 is placed on the interpolated curve between the points with the ordinates 73 and 51_5. The point with the ordinate 33 has then an abscissa at a distance from that with the ordinate value 55, which corresponds to the accepted "intensity ratio" of the pair. Using the characteristic curve constructed in this way, one obtains intensity ratios proportional to the true intensity ratios.[11] Accordingly, the analytical curves (intensity ratio—concentration relation) for the elements in the samples can be found from the spectrograms, applying for each plate the appropriate calibration.

5. SELECTIONS OF LINES

In connection with plate calibration one is confronted with the proper selection of lines of the necessary stability. It is of interest whether the measured intensities of these lines can be expected to be constant regardless of type of source, spectrograph, and densitometer used. The requirements on the constancy of the relative intensity of lines used for plate calibration must be very high.

It might seem that the crucial test for the constancy of intensity ratios is the comparison of the spectra in an arc and in a spark of some kind. The excitation, because of the difference in temperature, and reabsorption, because of different vapor density, are in these two sources so unlike, that if the lines stand this test, the same can be expected for any other type of excitation. This test is, however, not sufficient. Although lines with stable relative intensities are among those which stand this test, further variations of the circuit parameters may reveal that the lines are not suitable for calibration purposes, for use under changeable conditions. Many of the lines having practically the same relative intensities in the spark of certain parameters (high self-inductance, for instance) and in the arc, show somewhat different measured relative intensities when the parameters are changed, e.g., a small self-inductance is used or when the electrodes are of a different structure.

Several iron lines were investigated in this laboratory in that respect by variation of the self-inductance and capacitance in the spark and by using alternatively two steel pencils as electrodes, or pellets made of steel drillings and sparked against a graphite rod. These are the usual types of samples applied for steel analysis, and it is of importance whether the lines can be used for plate calibrations (or for determination of the contrast) on any spectrogram of a steel sample. Most of the lines which withstood the first-mentioned test showed noticeable, and some considerable, change of relative intensities when measured on spectrograms made under different sparking conditions or with different electrodes. A superficial inspection of the spectrograms reveals that the sharpness of the lines is different, being in general more diffuse in a spark with small self-inductance. Whether the relative intensity of the lines is actually changed or the maxima of the lines which are considered in the usual microphotometer technique as the measure of the line intensity are affected by the line broadening, or whether continua overlap the line spectra in different degree, has not yet been ascertained. Whatever the reasons, the possibility of a change in the (measured) intensity ratio must always be considered when sparking con-

[11] If the intensity ratio assigned to the gamma-pair is the true ratio, the calibration curve will yield true ratios only for wave-lengths in the immediate vicinity of the gamma-pair. For other wave-lengths the calibration curve will in general not yield true ratios anyhow. The fact that one calibration curve is often successfully used for working pairs widely scattered in the spectrum seems to indicate that the units of the log intensity axis of the characteristic curves for different wave-lengths are approximately proportional on plates of changeable contrast. Some direct determinations of the characteristic curves for different wave-lengths made on plates with different contrasts seem to support this conclusion. More work has to be done, however, in order to obtain conclusive results.

TABLE II.

Sample No.	Gamma-pair ratio of the plates measured with the basic plate calibration	Intensity ratios of different pairs of lines measured with the appropriate plate calibration Pair I	Pair II	Pair III	Emulsion
1	0.46	1.53	1.48		Sp. An. No. 1
	0.51	1.53	1.45		Sp. An. No. 1
	0.45	1.50	1.50		Sp. An. No. 1
	0.55	1.53	1.43		Infra-Red LI
	0.56	1.60	1.51		Infra-Red LI
2	0.44	1.56	1.88		Sp. An. No. 1
	0.50	1.49	1.94		Sp. An. No. 1
	0.44	1.50	1.83		Sp. An. No. 1
	0.50	1.57	1.91		Sp. An. No. 1
	0.53	1.47	1.89		Sp. An. No. 1
	0.56	1.51	1.83		Infra-Red LI
	0.55	1.52	1.87		Infra-Red LI
3	0.45	0.85	0.72	1.34	Sp. An. No. 1
	0.45	0.86	0.76	1.28	Sp. An. No. 1
	0.50	0.85	0.75	1.31	Sp. An. No. 1
	0.57	0.84	0.75	1.31	Sp. An. No. 2
	0.57	0.88	0.75	1.31	Sp. An. No. 2
4	0.48	1.14	1.21	1.76	Sp. An. No. 1
	0.56	1.15	1.23	1.83	Sp. An. No. 2

ditions are changed. The width of the spectrograph slit, as well as the type of the densitometer and the width of its slit, can also affect the measured ratios, for understandable reasons.

The intensity ratio of the lines Fe II 2805.8A and Fe II 2827.4A were found to be relatively stable in spark sources of different parameters. The intensity ratio of these two lines was found to be close to 1:2. The value 0.50 was assigned to this gamma-pair and a set of permanent calibration curves for different contrasts was derived from the basic curve in such a way as to obtain the ratio 0.50 with the blackenings of the gamma-pair lines on plates of different contrast, using the appropriate calibration. This set of curves covered the range of gamma-pair ratios from 0.42 to 0.58, as measured with the basic calibration curve.

The intensity ratio of another pair of iron lines, Fe I 3166.4 and Fe I 3165.9, with the ratio close to 0.60, was found to be also of satisfactory stability except for a somewhat larger change in the ratio (in comparison with the previous pair) when steel pellets against carbon as electrodes were used, instead of two steel pencils.

6. SOME EXPERIMENTAL RESULTS

In order to test the validity of the assumptions made for deriving the permanent calibrations and to check the applicability of the latter in practical spectrographic work, experimental characteristic curves were found for plates of different contrast. These curves coincided with the curves derived from the basic one, within the experimental errors. A more sensitive and practically important test is the comparison of the intensity ratio of a pair of lines measured on plates of different contrast, applying the appropriate derived characteristic curves. Eastman Kodak Spectrum Analysis No. 1 plates were available of different delivery dates. Those delivered more than a year ago had a contrast corresponding on the average to the gamma-pair ratio 0.50. The plates of a recent delivery had a ratio averaging 0.44. The contrast of the (developed) plates within each group and even within each batch varied in such a way that the gamma-pair ratio covered the whole range between 0.42 and 0.52. In addition, some plates of other emulsions were tested, namely, Spectrum Analysis No. 2 and Infra-Red LI, in order to obtain still lower contrast, although for these two emulsions a good agreement with the results obtained with the Sp. An. No. 1 plates was not to be expected, on account of the supposedly better uniformity of the contrast for different wave-lengths of the Sp. An. No. 2 plates and the sensitizing of the Infra-Red plates. Actually, the average results obtained with these plates deviated in the investigated range of transparencies (that is, from 0.10 to 0.80), from the average values of the No. 1 emulsions not much more if at all than those obtained with different plates of the same emulsion. The individual results on plates with lower contrast (higher gamma-pair intensity ratio) showed, naturally, a greater scattering. The No. 2 and LI emulsions had in the investigated spectral range a contrast corresponding to about 0.55–0.57 of the gamma-pair ratio. Eastman Kodak D-19 developer for most of the plates for three minutes at 65°F and occasionally, D-8 for 1.5 minutes were used.

In Table II there are some typical results given obtained with different emulsions. In the second column the gamma-pair ratio of the plate is given as measured with the basic calibration. In the columns I, II, III, the intensity ratios of various pairs of lines with wave-lengths between 2800 and 3300, measured with the appropriate calibration curve. Each value is an average of from 5 to 10 spectrograms on the same plate. Thus the occasional fluctuations of the light

source and possible non-uniformity of the developed plates did not affect appreciably the measured intensity ratios.

Numerous measurements with the same samples made on different plates, similar to those in Table II, showed an (arithmetical) average deviation from the mean values of about ±1.5 percent of ratios not exceeding 2:1. The closer to unity the ratio, the smaller the deviations, as the difference in plate calibrations becomes of little importance.

When several spectrograms are made on the same plate and the time permits, it is advisable to measure the gamma-pair ratio on several spectrograms and to apply the mean value. Also, different gamma-pairs, of different ratios, can be used for better determination of the plate contrast. In such cases the advantage compared with the usual method of plate calibration is not the elimination of numerous measurements but avoiding the plotting of the curve in haste, which can occasionally be a source of considerable error, particularly for higher densities.

In order to check the proper choice of the plate calibration, in addition to the measuring the gamma-pair ratio, the following procedure is often convenient, when the concentration can be determined by means of two different analytical pairs. In these cases the reference lines (internal standards) are chosen usually in such a way that the intensity ratio of one analytical pair is smaller than 1, that of the other pair is larger than 1. The concentration determined with two such analytical pairs is identical only when measured with the correct plate calibration curve, as otherwise the calculated intensity ratios are distorted in the opposite directions. The selection of the calibrations in the series reproduced in Table II was, however, not corrected by this method.

SUMMARY

The possibility of application of permanent plate calibrations for analytical spectroscopy is discussed and the preparation of a set of calibrations for plates of different contrast is described. Some experimental results are given illustrating the applicability of the procedure.

The writer expresses his sincere gratitude to the members of the Spectrographic Laboratory, Chemical Department, Gary Works for all the measurements made in connection with this investigation. He is particularly obliged to D. I. Byrge, O. W. Christine, R. H. Colin, and J. W. Macedo for their active help.

JOURNAL OF THE OPTICAL SOCIETY OF AMERICA VOLUME 34, NUMBER 8 AUGUST, 1944

Spectrographic Determination of Trace Elements in 70/30 Brass and Admiralty Brass. Part I*

P. A. LEICHTLE
Chase Brass & Copper Company, Inc., Waterbury, Connecticut

A spectrographic procedure for the simultaneous determination of Pb, Fe, Ni, Sn, Bi, and Sb in 70/30 brass and admiralty brass is described in detail. The source is a 14-ampere low voltage d.c. arc of conventional type. A 50-mg pellet sample is arced to extinction in a cupped graphite electrode of specified dimensions. The "intermediate mode" of illumination obtained by focusing the source on the collimator lens is used. A radial sector disk placed at the slit is used to regulate the effective exposure at the plate. The step sector method of plate calibration is used. The source for the calibration is a 4-ampere d.c. arc on $\frac{3}{16}$" diameter copper electrodes. The calibration curve on the copper line 2882.934A is used for a wave-length region 2590A–3100A. The copper reference line of 2858.734A is chosen for comparison with the respective impurity lines. Working curves are given for the determination of Pb, Fe, Ni, Sn, and Sb in the range of 0.01–0.10 percent and Bi in the range of 0.0004–0.010 percent.

INTRODUCTION

THE Analytical Section of our Research Department has the responsibility of testing a large number of copper and brass products for conformity with specifications of composition. The chief chemist has various analytical tools and methods at his disposal. Generally, the major constituents are analyzed by chemical

* Presented at the Optical Society Meeting, October 7–9, 1943, at Pittsburgh, Pennsylvania.

methods, while trace elements and some of the minor constituents are determined by spectrographic methods.

One of the important copper alloys at this time is cartridge brass or "70/30 brass." It is the accepted grade of deep drawing brass for the manufacture of cartridge cases. The chemical specification places a limiting value of 0.15 percent total of elements other than copper and zinc. The permissible lead and iron content is also specified. Admiralty brass is also of considerable importance. It is widely used in the form of condenser tubes. The copper content of the two alloys is similar. A summary of the American Society for Testing Materials specifications 29 for these two alloys is given in Table I.

In this discussion, special attention will be given to the spectrographic determination of lead in cartridge brass. If the cartridge brass casting is to be hot rolled, the brass manufacturer must keep the lead content under 0.010 or 0.015 percent. This requires careful analytical control.

An excellent variety of spectrographic methods for copper and copper alloys has been published. Very few investigators appear to favor spark excitation methods, although Lomakin[1] has suggested a spark source for the determination of bismuth in copper. Kaiser[2] states that any element having a high work function; i.e., iron platinum, copper, and silver, gives irregular types of discharge with spark excitation.

Sventitskii[3] suggests an a.c. arc source for the spectrographic analysis of brass and bronze. Gerlach and Riedl[4] advise the use of an intermittent arc for analysis of trace elements in pure metals rather than a continuous arc which is said to give erroneous results due to selective vaporization of low boiling point elements. Ratsbaum[5] applied this intermittent arc method to the approximate determination of arsenic, antimony, bismuth, tin, lead, and iron in electrolytic copper.

Breckpot[6] stated that fractional distillation phenomena of the more volatile constituents complicate the quantitative spectrographic analysis of copper by the electric arc method. These selective effects were said to be eliminated by examining the sample as the cathode. He prepared analytical curves for lead, bismuth, silver, arsenic, and antimony in copper by his method.[7] Representative average values were obtained on copper oxide powders prepared from appropriate solutions.

For the spectrographic analysis of brass, Brownsdon and Van Someren[8] favor a self-electrode method and d.c. arc excitation. The lower self-electrode is made the cathode and the counter electrode is a pure copper rod. Further applications of this method were described by Smith;[9] and also by Barker.[10]

The d.c. arc excitation of copper or copper alloy globules has been described by several investigators. Hill and Luckey[11] described a visual spectroscopic method for lead in copper. A 10-ampere d.c. arc source was used on a 0.4-gram sample of copper supported in a cupped graphite electrode. The percentage of lead was correlated with the time of visibility of the lead line 4058A.

TABLE I.

Cartridge brass		
	Copper, percent	68.50 to 71.50
	Lead, max. percent	0.05
	Iron, max. percent	0.05
Total of elements other than copper and zinc,	max. percent	0.15
	Zinc, remainder	

Note: The presence of bismuth in excess of 0.006 percent may constitute cause for rejection of cartridge brass disks or strip.

Admiralty brass		
Type A:	Copper, percent	70.00 to 73.00
	Tin, percent	0.90 to 1.20
	Lead, max. percent	0.075
	Iron, max. percent	0.060
	Zinc, remainder	

Type B is Type A modified by the addition of arsenic—max. percent 0.10

Type C is Type A modified by the addition of antimony—max. percent 0.10

Type D is Type A modified by the addition of phosphorus—max. percent 0.10

[1] B. A. Lomakin, Zeits. f. anorg. allgem. Chemie **187**, 75–96 (1930).

[2] H. Kaiser, *Proceedings of the Seventh Summer Conference on Spectroscopy and Its Applications* (John Wiley & Sons, Inc., New York, 1940), pp. 77–81.

[3] N. S. Sventitskii, Zavodskaya Lab. **7**, 1371–8 (1938).

[4] We. Gerlach and E. Riedl, Metallwirtschaft **12**, 401–5 (1933).

[5] E. A. Ratsbaum, Zavodskaya Lab. **6**, 191–5 (1937).

[6] R. Breckpot, Chim. et Ind. **31**, 220–9 (1934).

[7] R. Breckpot, Ann. Soc. Sci. Bruxelles **54**, 299–314 (1934).

[8] H. W. Brownsdon and E. H. S. Van Someren, J. Inst. Metals **46**, 97–113 (1931).

[9] D. M. Smith, *Metallurgical Analysis by the Spectrograph* (British Non-Ferrous Metals Research Association, London, 1933).

[10] F. G. Barker, J. Iron Steel Inst. (London) **139**, 211–55 (1939).

[11] C. W. Hill and G. P. Luckey, Trans. Am. Inst. Min. Met. Eng. **60**, 342–353 (1919).

Milbourn[12] also used a globule method for the spectrographic determination of minute quantities of impurities in copper. A 0.2- to 0.5-gram sample was supported in a shallow graphite cup. In this instance, the sample was made the cathode, while the counter electrode was a pure copper rod. Milligan and France[13] analyzed phosphor bronze for phosphorus by a globule method. A 0.2-gram sample, supported in a graphite cup, was arced in a 10-ampere d.c. arc for 60 seconds. The sample was made the anode and the counter electrode was a pointed graphite rod. France[14] also described a similar procedure for the determination of impurities in cupro-nickel. A 0.2-gram sample was arced at 10 amperes for 30 seconds. Harrison[15] illustrated the variation in the rate of volatilization of different constituents in a d.c. arc by making a moving plate spectrum of a 50-mg brass sample. He emphasized that great care must be employed in the use of a d.c. arc to insure identical treatment of the unknown and the standard samples.

Several papers of Mott[16,17] give much useful information on the behavior of various elements in the carbon arc. He stated that metals which form molten beads and which do not wet or dissolve the carbon will give arcs in which the temperature and distillation conditions are more constant and uniform than is the case for metals wetting the carbon or for irregular partially molten masses. (Most of the copper alloys fall within the first classification.)

Various factors affecting the operation of a carbon arc were described by Chaney, Hamister, and Glass;[18] and MacPherson.[19] Conditions which give a reproducible maximum temperature at the positive crater of a pure carbon arc were specified.

Combination chemical and spectrographic methods for the determination of minute impurities in copper have been described by Breckpot,[20] Park,[21] and Park and Lewis.[22] Increased sensitivity was obtained by concentration of the impurities by chemical separation of the bulk of the copper and analyzing the concentrate by spectrographic methods.

The American Society for Testing Materials Committee E-2 on Spectrographic Analysis has done considerable work on copper alloys, with the emphasis on specification testing for trace elements. A wide variety of d.c. arc methods for such alloys has been proposed by various members of the committee.

In the Research Department of the Chase Brass & Copper Company, we have done a great deal of experimental work on spectrographic methods for copper alloys. Early experiments with a simple condensed spark circuit of 1-kva input indicated that this source was unsuitable for detection of impurities in 70/30 brass. Similar tests with a commercial Feussner spark apparatus were also discouraging because of poor sensitivity.

Various d.c. arc methods have been tested. A modification of the self-electrode method of Brownsdon and Van Someren was described by the author.[23] We used a $\frac{1}{8}$" diam. self-electrode of the brass and a counter electrode of $\frac{3}{16}$" diam. graphite rod. A current of 3 amperes was used and the self-electrode was the cathode. An average precision of ± 12 percent was reported for the determination of antimony in admiralty brass (type C). We have also found this method to be useful in identification and control testing of many other types of copper alloys. However, it is not a universal method because of the difficulty in obtaining self-electrodes from many items of finished brass mill products such as, for example, thin sheet and fine wire.

In nearly every instance, both chemical and spectrographic tests are required. Obviously, there would be a considerable saving in time and labor if millings, drillings, or clippings could also be used for the spectrographic tests. We have adopted such a method for the spectrographic determination of trace elements in many of our

[12] M. Milbourn, J. Inst. Metals **55**, 275–82 (1934).
[13] Wm. E. Milligan and Walter D. France, Ind. Eng. Chem. Anal. Ed. **32**, 24–27 (1941).
[14] Walter D. France, J. Opt. Soc. Am. **32**, 681–685 (1942).
[15] G. R. Harrison, Metals and Alloys **7**, 290–296 (1936).
[16] Wm. Roy Mott, Trans. Am. Electrochem. Soc. **31**, 365–390 (1917).
[17] Wm. Roy Mott, Trans. Am. Electrochem. Soc. **37**, 665–706 (1920).
[18] N. K. Chaney, U. C. Hamister, and S. W. Glass, Trans. Am. Electrochem. Soc. **67**, 201 (1935).
[19] H. G. MacPherson, J. Opt. Soc. Am. **30**, 189–194 (1940).
[20] R. Breckpot, Ann. Soc. Sci. Bruxelles **B55**, 173–194 (1935).
[21] B. Park, Ind. Eng. Chem. Anal. Ed. **6**, 189–190 (1934).
[22] B. Park and E. J. Lewis, Ind. Eng. Chem. Anal. Ed. **7**, 182–3 (1935).
[23] P. A. Leichtle, *Symposium on Spectrographic Analysis*, A.S.T.M. Bull. PP 49 (March, 1941).

copper alloys. Only a small portion of the chemical sample is required for the spectrographic test. The remainder of this discussion will deal chiefly with the application of this method to the simultaneous determination of six trace elements in 70/30 and admiralty brass. An alternative method for the determination of lead, only, will also be discussed.

I. APPARATUS

A Hilger quartz spectrograph of the large Littrow type (E-384) was used in all of this work. The slit width was 30 microns and the slit length was 1 mm. The collimating lens was used at full aperture.

The optical bench was fitted with a Hilger arc and spark stand; and a Jarrell-Ash step sector unit comprising a step sector disk (interchangeable with a radial sector disk), a quartz lens, and an adjustable speed motor. The quartz lens in the sector unit images the source on the collimating lens to give the "intermediate mode" of illumination. This was described by Breckpot and Lialine,[24] and also was used by Brode.[25] A uniform optical alignment of the arc source was obtained by a separate auxiliary lens in a stand offset from the center line of the optical bench. This lens images the source on a target which was attached to the box of the spectrograph seven inches to the right of the slit.

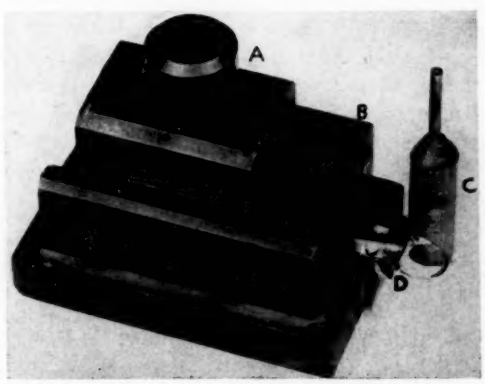


FIG. 1. Photograph of the pellet press used in compressing the weighed sample of brass millings or drillings into circular disks $\frac{3}{16}$" diam. *A*, mold; *B*, supporting anvil; *C*, punch; *D*, receptacle. With the mold in position to the right, the weighed sample is brushed into the mold, the punch is inserted and lightly tapped with a copper hammer. The mold is then moved to the left position (as shown in the photo) and the disk sample drops through an opening in the supporting anvil into a small drawer-like receptacle.

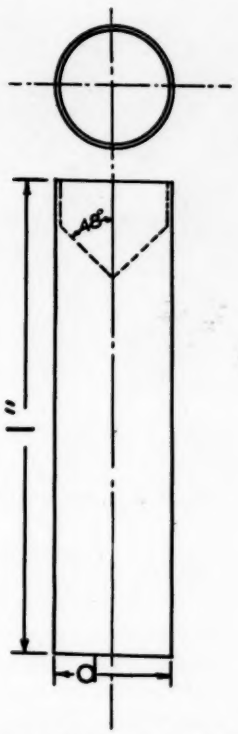


FIG. 2. Diagram of the cupped graphite electrodes used in the spectrographic testing of 70/30 brass and admiralty brass. *d*—diam. of graphite rod = 0.250"; drill angle = 48°; cup depth = 0.19"; and inside diam. = 0.215".

The arc source was supplied by a 220-volt d.c. power line. The arc current was controlled by means of a carbon-pile resistor unit and was measured on an ammeter, 0–25 scale. The voltage drop across the arc was measured by a d.c. voltmeter, 0–250 scale.

The Eastman Spectrum Analysis No. 1 plates were used in all this work. Developer temperature was kept constant at 70°F ± 0.5° by a Kellogg-American tray cooler. A wide camel's-hair brush was used for brush development.

Transmission measurements on the selected lines were made with a Hilger non-recording microphotometer. The 18-watt projection bulb was operated at 2.8 amp. and the power source

[24] R. Breckpot and L. Lialine, Ann. Soc. Sci. Bruxelles **54**, 299–314 (1934).

[25] W. P. Brode, *Symposium on Spectrographic Analysis* (A.S.T.M., 1935), Vol. 35, Part II, pp. 1–6.

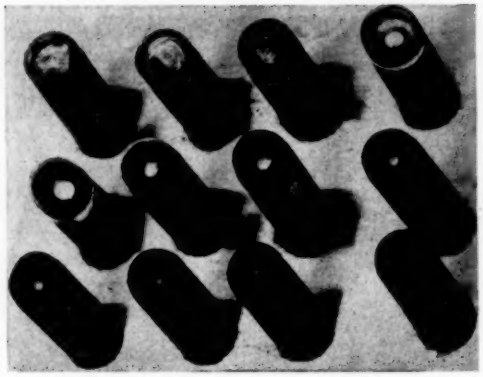


FIG. 3. A photograph showing the appearance of the globule and supporting graphite electrode during an average exposure. Reading from left to right: back row: at 0, 2, 3, 5 seconds; middle row: at 10, 20, 30, 40 seconds; front row: at 50, 60, 70, 80 seconds, respectively.

was two automobile lead storage batteries connected in parallel.

The brass samples for the spectrographic tests were weighed on a student type chemical balance (sensitivity $-\frac{1}{10}$ mg). The weighed brass chips were placed in the mold of a small pellet press (Fig. 1) and compressed to a $\frac{3}{16}''$ diam. flat disk.

The regular grade of graphite as supplied by the National Carbon Company was found suitable for use as supporting electrode for the testing of brass samples. A Jarrell-Ash graphite driller was used to prepare the cupped electrodes from $\frac{1}{4}''$ diam. graphite rod. Figure 2 gives the specifications for the ideal cupped electrodes. In actual practice, the cup depth and the drill angle can easily be duplicated. A slight variation in wall thickness can be tolerated, but a uniform wall is extremely desirable. The best cupped electrodes were prepared by shaving an oversize rod (0.255″ or over) to 0.250″ outside diam. and drilling in one operation with the graphite driller mentioned above.

A calculating board of conventional design was provided for converting transmission values to relative intensities by means of a step sector calibration curve.

II. PREPARATION OF SAMPLES AND ANALYTICAL PROCEDURE

During the casting operation, a ladle sample for analytical testing is obtained from each furnace. Each ladle sample is milled on a commercial milling machine using a milling cutter 6″ diam. and $\frac{3}{16}''$ wide. This milled sample is used for both chemical and spectrographic tests. The milled sample usually needs no cleaning; however, before weighing the sample, it is magnetized to remove any possible contamination due to minute particles of iron introduced during the milling operation.

Wrought materials such as cartridge brass sheets, strips, or disks may be sampled by drilling, milling, sawing, or clipping. Admiralty brass tubes are sampled on a special milling machine so as to mill the entire cross section of the tube.

Standard samples were required for the preparation of analytical curves. These standards were prepared by melting the required alloying constituents in a small high frequency electric furnace, casting in flat bars, and processing to a final thickness of 0.144″. Each standard sample was sampled by milling and then was analyzed by approved chemical methods.

For the spectrographic tests, 50-mg samples were weighed on a chemical balance. The weighed chips were compressed in the pellet press to give flat disks $\frac{3}{16}''$ diam. and approximately 0.05″ thick. The disks were placed in prepared cupped $\frac{1}{4}''$ graphite electrodes and were arced to completion with d.c. arc excitation at 14 amperes. The upper electrodes were pointed $\frac{3}{16}''$ diam. graphite rods. An arc gap of 5 mm was maintained during the entire exposure. The radial sector at the slit was adjusted to give a transmission of 6 percent for the normal exposure.

The sample at various stages of the exposure is pictured in Fig. 3. The sample melts to a molten bead during the first 3 seconds. Most of the zinc (and traces of cadmium if present) volatilizes during the first 15 seconds. After 30 seconds of exposure, the remainder of the zinc, the lead, and the bismuth (if present) will have volatilized. The copper and the trace elements, iron, nickel, tin, antimony, arsenic, manganese, phosphorus, etc., are volatilized at nearly the same rate during the period from 30 seconds to completion. (This was verified by microphotometer studies of "moving plate" spectra.) The voltage drop across the arc is 30–32 volts during the period from 15 seconds to completion.

A slit length of 1 mm was used. This was the

same as is used in daily control. This allows 60 spectra and a step sector calibration spectrum to be recorded on each 4″×10″ plate. The calibration source was a 4-ampere d.c. arc on $\frac{3}{16}$″ diam. copper rods. The electrodes were preburned 10 seconds and the exposure time was 10 seconds. The optical arrangement remains the same as for the analytical spectra. The log step sector with 8 steps of step exposure ratio of 1.585 to 1 is substituted for the radial sector at the slit. In daily control testing of 70/30 brass and admiralty brass, it is often more convenient to use limiting standards on each plate than to put a step-sector calibration pattern on every plate. (Many of the spectra require only visual examination because the impurity content of the average sample seldom exceeds 35 percent of the permissible maximum.)

The Spectrum Analysis No. 1 plates were processed in Eastman D-19 developer, 2½ minutes at 70°F, using a wide camel's-hair brush. The plates were brushed gently at a rate of 12 times per minute. (Clark[26] gave a complete discussion in the technique of uniform development.) After development, the plates were agitated 30 seconds in an acetic acid stop bath, fixed 3 minutes in Eastman F-1 fixing bath, washed 10 minutes, and dried in a current of warm air.

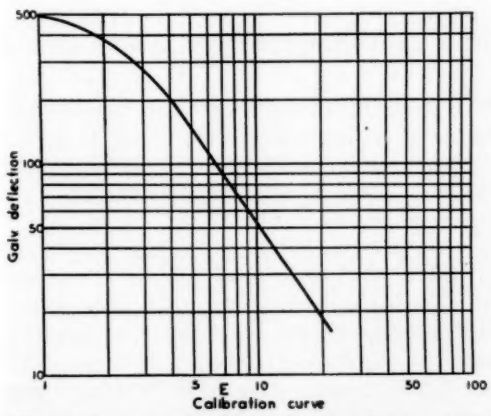


FIG. 4. Average calibration curve for the wave-length region 2590 to 3100A. Step sector of eight steps, step ratio 1.585 to 1. Source: 4-ampere d.c. arc on copper electrodes. Eastman Spectrum Analysis No. 1 plate. Brush development in D-19, 2.5 minutes at 70°F.

[26] Walter Clark, Photo Technique, pp. 54–58 (Oct. 1940).

TABLE II. Analytical line pairs.

Element line	Reference line	Equal density (%)	Relative sensitivity
Pb 2833.069	Cu 2858.734	0.011	200
2873.316	Cu 2858.734	0.071	31
Fe 2966.900	Cu 2858.734	0.021	105
Ni 3050.819	Cu 2858.734	0.011	200
3054.316	Cu 2858.734	0.039	56
Sn 2839.989	Cu 2858.734	0.006	370
2863.327	Cu 2858.734	0.015	146
2850.618	Cu 2858.734	0.16	14
Sb 2598.062	Cu 2858.734	0.06	37
2877.915	Cu 2858.734	0.14	16
Bi 3067.716	Cu 2858.734	0.0022	1000
As 2349.84	Cu 2858.734	[a]	12
P 2535.65	Cu 2858.734	[a]	11

[a] No data.

III. ANALYTICAL CURVES AND INTERPRETATION OF DATA

Many useful spectral lines of iron, lead, tin, nickel, antimony, bismuth, manganese, silicon, and aluminum are found in the region 2590 to 3100A. The copper line 2882.934A was selected as the calibration line for this wave-length region. The calibration curves were plotted on log-log paper as suggested by King.[27] The galvanometer deflections were plotted *versus* exposure times of the respective sector steps (see Fig. 4).

The effective exposure at the plate, as controlled by the radial sector, was of such a value as to give line densities of 1.0 or less for most of the selected lines. The copper line 2858.734A was selected as the internal standard reference line. It is located centrally in the wave-length region, and is of suitable intensity for comparison with the selected trace lines.

The analytical line pairs used in this investigation are given in Table II (wave-length values by Harrison[28]). The equal density values, as given in column 3, were taken from the analytical curves. Relative sensitivities (column 4) were obtained by assigning an arbitrary sensitivity of 1000 to the bismuth line 3067.716A, and com-

[27] Carl King, J. Opt. Soc. Am. **32**, 112–115 (1942).

[28] G. R. Harrison, *Massachusetts Institute of Technology Wavelength Tables, with Intensities in Arc, Spark or Discharge Tube* (John Wiley & Sons, Inc., New York, 1939).

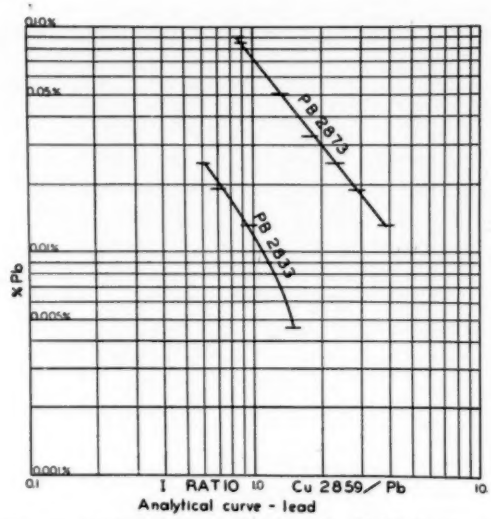


FIG. 5. Analytical curve for lead in 70/30 brass and admiralty brass.

paring each value in column 3 to the bismuth value in column 3. Spectra from a special sample of admiralty brass containing equal amounts of antimony, arsenic, and phosphorus were used to calculate the relative sensitivities of arsenic and phosphorus as compared to antimony.

The analytical curves for the analysis of lead, iron, nickel, tin, and bismuth were obtained from spectra of cartridge brass standards. Admiralty brass standards containing antimony were used for the antimony study.[29] (Figures 5–9 inclusive.) Tests on miscellaneous samples of admiralty brass containing known amounts of lead, iron, and nickel indicate that the cartridge brass data can be used for the analysis of either cartridge or admiralty brass for these elements. The analytical curve for bismuth is included because many specifications do mention that the presence of bismuth in cartridge brass is objectionable. However, bismuth is not normally found in cartridge brass in amounts exceeding 0.0003 percent.

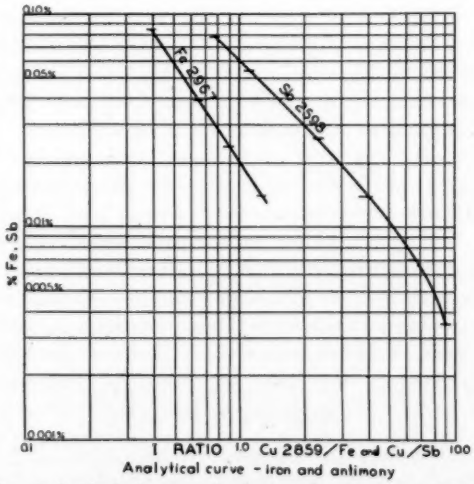


FIG. 6. Analytical curves for iron and antimony in 70/30 brass and admiralty brass.

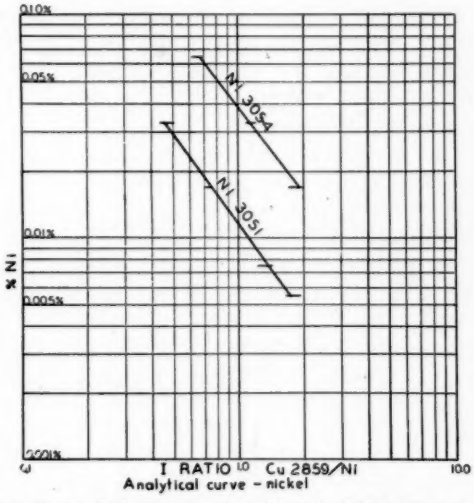


FIG. 7. Analytical curve for nickel in 70/30 brass and admiralty brass.

In preparing the spectra of the standard alloys, many of the standards were arced at several different sector transmissions in order to obtain the most reliable microphotometer data. The higher standards on (1) the Fe 2967 curve, (2) the Ni 3051 curve, (3) the Sn 2863 curve, and (4) the Bi 3068 curve were exposed at a sector-transmission of normal and also at one-half normal to reduce the density of the trace lines. The lower standards on (1) the Pb 2873 curve, (2) the Sb 2598 curve, and (3) the Sn 2851 curve were exposed at normal and also at 1.5 times normal transmission to increase the density of the impurity line. The normally exposed spectra (at sector transmission of 6 percent) gave average back-

[29] *Standards for Copper and Copper Alloys* (American Society for Testing Materials, Philadelphia, 1942), pp. 60–65.

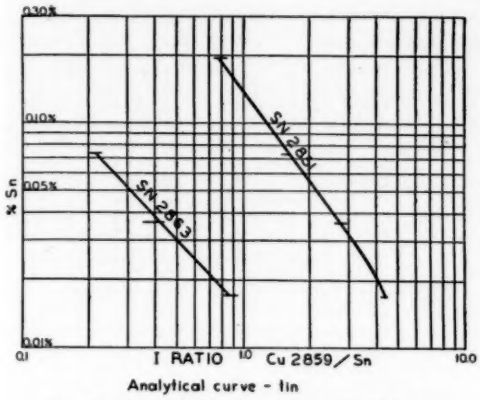


FIG. 8. Analytical curve for tin in 70/30 brass.

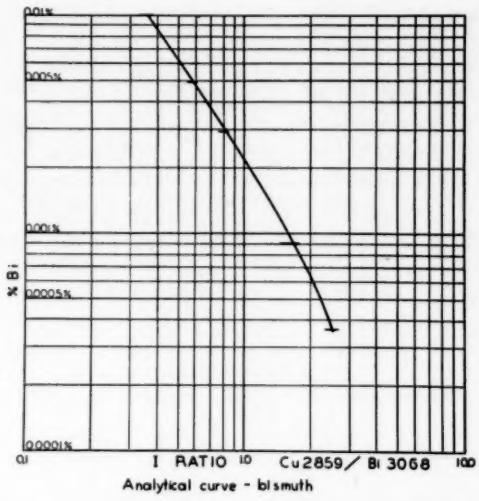


FIG. 9. Analytical curve for bismuth in 70/30 brass.

ground densities of 0.02 density. This was neglected in calculating the relative intensities of a line pair.

The use of this method for the analysis of minor constituents such as tin in admiralty brass is being studied. No data will be given here. (However, this same method has been applied to the semi-quantitative analysis of tin in copper-tin alloys in the range of 0.5 percent, 1 percent, 3 percent, and 5 percent tin. The line pairs Cu 2859/Sn 2851, Cu 2859/Sn 2594, and Cu 2859/Sn 2913 were studied.)

A quantitative measure of the relative volatilization rates of the trace elements in cartridge brass and admiralty brass is given in Table III. Columns 3, 4, and 5 give the apparent composition of the material voltatilized during the period 0–45 seconds, 0–60 seconds, and 0–80 seconds, respectively. The results indicate that lead and copper have wide differences in volatilization rates, while there is little practical difference in volatilization rates of the trace elements iron, nickel, antimony, and tin as compared to the matrix element copper.

A logical alternative method for the determination of lead would be to add to the sample in the cup a known weight of an internal standard reference element having nearly the same boiling point as lead. Bismuth was selected as that element. It was added as a copper-bismuth alloy containing 0.019 percent bismuth. This alternative method specifies the addition of 50

TABLE III.

Sample	Chem. anal.	Apparent composition 45 seconds exp.	60 seconds exp.	80 sec. (completion)
169	0.025 Pb	0.070–0.047–0.046	0.033–0.036–0.037	0.026–0.028–0.028
170	0.033 Pb	0.068–0.090–0.088	0.042–0.036–0.047	0.034–0.035–0.033
171	0.051 Pb	0.126–0.130–0.115	0.068–0.057–0.078	0.047–0.044–0.051
169	0.014 Fe	0.013–0.014–0.013	0.014–0.012–0.012	0.014–0.013–0.015
170	0.024 Fe	0.018–0.022–0.021	0.019–0.019–0.018	0.022–0.023–0.022
171	0.039 Fe	0.035–0.034–0.035	0.031–0.032–0.034	0.042–0.039–0.042
169	0.0075 Ni	0.0062–0.0067–0.0069	0.0060–0.0060–0.0060	0.0080–0.0080–0.0090
170	0.017 Ni	0.013–0.014–0.012	0.012–0.012–0.012	0.017–0.019–0.018
171	0.033 Ni	0.023–0.025–0.025	0.022–0.023–0.024	0.033–0.032–0.034
25	0.073 Sn	0.068–0.067	0.071–0.073	0.073–0.073–0.070
210	0.014 Sb	0.012–0.011–0.013	0.012–0.012–0.013	0.015–0.015–0.014
50	0.039 Sb	0.035–0.034–0.036	0.036–0.038–0.035	0.042–0.043–0.043
52	0.078 Sb	0.066–0.071–0.070	0.078–0.080–0.070	0.078–0.080–0.070
192	1.24 Sn	1.10–0.96–1.06	1.06–1.08–1.25	1.23–1.18–1.30
73	2.82 Sn	2.55–2.45–2.60	2.30–2.70–2.60	2.85–2.80–2.85
57	5.15 Sn	3.60–3.90–3.60	4.10–3.70–4.20	4.80–5.00–5.00

TABLE IV.

Sample	Chem. anal.	Apparent composition 10 amp.	12 amp.	14 amp.	16 amp.
374	0.054 Sb	0.054–0.053–0.054	0.054–0.053–0.051	0.055–0.053–0.053	0.051–0.052–0.053
23	0.017 Sn	0.022–0.020–0.021	0.019–0.019–0.019	0.018–0.019–0.018	0.018–0.017–0.018
25	0.073 Sn	0.077–0.074–0.072	0.073–0.070–0.072	0.070–0.069–0.073	0.072–0.070–0.068
150	0.0009 Bi	0.0014–0.0011–0.0011	0.0009–0.0010–0.0011	0.0009–0.0009–0.0008	0.0009–0.0009–0.0009
199	0.013 Pb	0.014–0.013–0.014	0.014–0.012–0.013	0.014–0.016–0.014	0.013–0.012–0.015
169	0.025 Pb	0.033–0.028–0.029	0.025–0.027–0.023	0.026–0.025–0.024	0.027–0.026–0.027
170	0.033 Pb	0.037–0.041–0.035	0.040–0.038–0.033	0.034–0.038–0.034	0.036–0.036–0.036
171	0.051 Pb	0.051–0.052–0.048	0.048–0.051–0.049	0.052–0.052–0.047	0.056–0.049–0.051
169	0.014 Fe	0.017–0.015–0.016	0.015–0.014–0.014	0.015–0.014–0.014	0.014–0.016–0.015
170	0.024 Fe	0.026–0.026–0.025	0.026–0.023–0.025	0.024–0.023–0.024	0.025–0.024–0.024
171	0.039 Fe	0.042–0.041–0.042	0.036–0.039–0.036	0.039–0.038–0.037	0.039–0.040–0.042
169	0.0075 Ni	0.0074–0.0068–0.0068	0.0074–0.0068–0.0071	0.0075–0.0077–0.0072	0.0075–0.0081–0.0073
170	0.017 Ni	0.018–0.018–0.018	0.017–0.016–0.016	0.018–0.016–0.018	0.016–0.017–0.017
170	0.017 Ni	0.019–0.018–0.018	0.018–0.016–0.018	0.018–0.016–0.017	0.019–0.018–0.018
171	0.035 Ni	0.033–0.036–0.036	0.029–0.033–0.028	0.030–0.036–0.033	0.037–0.038–0.031
192	1.25 Sn	1.28–1.23–1.22	1.17–1.17–1.20	1.22–1.21–1.22	1.13–1.15–1.21

TABLE V.

Sample	Chem. anal.	Apparent composition (1)	(2)	(3)
374	Sb 0.054	0.056–0.055–0.054	0.055–0.056–0.052	0.055–0.053–0.050
25	Sb 0.073	0.082–0.082–0.060	0.082–0.075–0.073	0.073–0.078–0.078
171	Pb 0.051	0.052–0.052–0.054	0.035–0.042–0.039	0.037–0.039–0.045
171	Fe 0.039	0.038–0.039–0.039	0.040–0.045–0.039	0.037–0.039–0.039
171	Ni 0.033	0.032–0.032–0.032	0.034–0.034–0.035	0.032–0.031–0.034

(1) Regular determination at 14 amperes.
(2) Arc current varied by steps from 10–12–14–16 during each exposure.
(3) Arc current varied irregularly between 12 and 16 amperes.

mg of the Cu-Bi alloy to each of the 50-mg cartridge brass samples. For this investigation, the 100-mg composite samples were arced for 1 minute at 14 amperes. Standard sized graphite cups were used, and the normal exposure was 6 percent sector transmission. The bismuth line at 2897.975A was selected as the reference line and the lead lines 2833.069A and 2873.316A were the selected trace lines. The analytical curves (Fig. 10) were prepared from selected standard brasses of 60 percent, 70 percent, 80 percent, and 85 percent copper content, and lead content from 0.01 to 0.15 percent.

Table IV illustrates the results obtained by arcing at several different current values. Fourteen cartridge brass samples, one admiralty brass sample (V374), and one copper tin sample (No. 192) were arced in triplicate at 10, 12, 14, and 16 amperes, respectively. In this range of current values there appears to be no measurable shift in relative intensities of the selected line pairs.

The effect of irregular variation in arc current during the exposure is illustrated in Table V. Column (1) gives the results obtained by the regular determination at 14 amperes. Column (2) gives the spectrographic results obtained by varying the current during each exposure in the following manner: 10 amperes, 0–30 seconds; 12 amperes, 30–45 seconds; 14 amperes, 45–60 seconds; 16 amperes, 60–80 seconds. There appears to be little or no effect on the antimony,

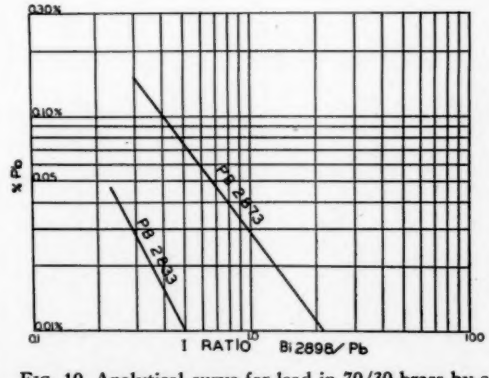


FIG. 10. Analytical curve for lead in 70/30 brass by an alternative method, using bismuth as the internal standard reference element.

tin, iron, and nickel results, but the lead results are low. Column (3) gives the results when the arc current is varied in an irregular manner in the range from 10 to 16 amperes. Again the Pb results are low. It is evident that the most reliable lead results will be obtained by maintaining the arc current at a constant value during the entire exposure.

IV. DISCUSSION

Extensive tests for the purpose of calculating the precision of the method have not been conducted. However, a study of the data used in plotting the analytical curves indicates the probable maximum deviations of pairs of values are as follows:

Lead	±10 percent (at a concentration of 0.05 percent)
Iron	±10 percent (at a concentration of 0.05 percent)
Nickel	± 9 percent (at a concentration of 0.05 percent)
Tin	± 7 percent (at a concentration of 0.05 percent)
Antimony	± 6 percent (at a concentration of 0.05 percent)
Bismuth	±10 percent (at a concentration of 0.005 percent).

Wider deviations may be encountered in routine testing unless extreme care is taken to avoid contamination of the sample with millings from other copper alloys. Contamination may cause the sample to exceed the limits, in which case a retest on a new sample is required. The conclusive spectrographic test is made on a sample from the corresponding cake, billet, or flat bar. The elements other than lead are rechecked in duplicate by the regular method, while the lead is rechecked by the alternative method for lead (Fig. 10).

The d.c. arc methods described in this paper have given satisfactory results for the simultaneous determination of six trace elements in cartridge brass and admiralty brass. The saving of time is an important advantage over chemical methods. The ease of sampling is an important advantage over spectrographic methods requiring self-electrodes. Two operators working as a team will normally analyze 120 samples in a 4-hr. period. This includes preparation of samples, exposure, processing of the photographic pates, necessary photometry, and calculations.

A minimum of manipulative technique is required of the operator, and comparable results are obtained among several operators. The use of a microphotometer eliminates personal errors in visual estimation of relative densities of line pairs.

The method can be adapted to the qualitative and quantitative listing of a wide variety of copper alloys. The binary brasses from 58 to 95 percent copper content, phosphor bronze, leaded copper, tellurium copper, cadmium bronze, leaded brass, and various copper-base bronzes may be listed as suitable for testing by the spectrographic procedure described herein. The average spectrographic laboratory would probably be equipped to use this method.

A continuance of this report on the determination of other trace elements in 70/30 brass and admiralty brass is being prepared. This may include such elements as manganese, arsenic, phosphorus, cadmium, silicon, and aluminum. (If the general method, as described herein, is being used, the presence of 0.01 percent or more of silicon and/or aluminum will invariably cause the pellet to be expelled from the cup during the early part of the exposure.)

ACKNOWLEDGMENTS

The author is indebted to Mr. A. Wickrowski for invaluable assistance in collecting the data for this paper, to Dr. H. L. Burghoff for preparation of standard samples, and to Mr. E. L. Smith for chemical analysis of the standards. Thanks are also due to Dr. D. K. Crampton and Mr. B. H. McGar for their interest in this investigation and to the Chase Brass & Copper Company for permission to publish this paper.

Dark Adaptation: Some Physical, Physiological, Clinical, and Aeromedical Considerations*

CHARLES SHEARD
Director, Division of Physics and Biophysical Research, Mayo Clinic and Mayo Foundation, Rochester, Minnesota

IN the last few years there has been accumulated by physiologists, biochemists, and biophysicists a considerable amount of very precise and, in some instances, new data and information concerning various visual functions. Recent measurements and evaluations give much support to the *duplicity theory*—a theory suggested by Max Schultze in 1866, independently suggested by Parinaud in 1881 as the result of his researches on night blindness, and finally correctly employed by von Kries in 1895 to explain the many properties and ramifications of the Purkinje phenomenon and to establish the double function of the retina in terms of the rods and cones.

I. BIOPHYSICAL AND PHYSIOLOGICAL ASPECTS

The retinas of most vertebrates contain two different types of receptors—cones and rods. Hence the retina of the vertebrate may be considered as a dual rather than a single sense organ. Structurally the center of the human eye, the fovea centralis, is occupied exclusively by cones, whereas the remainder of the retina contains cones and rods, with rods becoming increasingly predominant as the survey is made from center to periphery. The retinal topography of cones and rods is shown in Fig. 1. The most efficient vision under high illuminations is at the center (macula) of the retina, whereas at low illuminations the peripheral areas are more effective and serviceable than the center. Hence the idea of a dual visual function associates, on the one hand, vision under high illuminations with the cones and, on the other hand, vision under low intensities with the rods. Furthermore, since color is discerned best at high intensities and not at all at very low intensity of light, the cones may be considered as specific receptors for color and the rods as general receptors for light only. Cones, therefore, are concerned primarily with form (acuity) and color; rods with the sense of light.

1. Spectral Sensitivity

At low intensities of illumination the *visibility curve* of the eye, or the relationship between the wave-length of light and the relative energy necessary to produce a specific effect, is determined by the rods because the cones do not function, whereas at high intensities of illumination the visibility curve is determined by the cones because of their numerical preponderance in the fovea (Abney and Festing, 1891; Rosenberg, 1928, and others). The response of the visual receptors at intermediate intensities in retinal locations which have rods and cones depends on whether the contributions from the two systems remain separate or are combined into the resulting sensory effect. The situation is fairly simple and straightforward when the considerations concern *brightness*. In 1891 Koenig and Ritter showed that, for the human eye, the relative brightness of the spectrum at different intensities passes gradually from one system to the other and that at some intermediate intensities the visibility of the spectrum shows two maxima (Sloan, 1928; Forbes, 1929). Hence brightness is a function in which the two systems, the cones and the rods, contribute their effects additively.

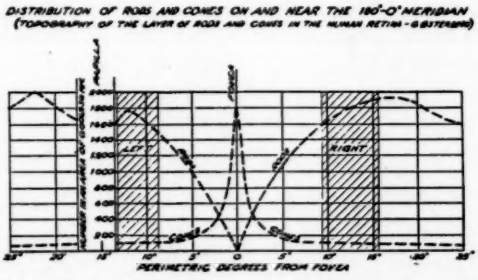

DISTRIBUTION OF RODS AND CONES ON AND NEAR THE 180°-0° MERIDIAN
(TOPOGRAPHY OF THE LAYER OF RODS AND CONES IN THE HUMAN RETINA - GØSTERBERG)
RODS
CONES
LEFT
RIGHT
BLIND SPOT
PERIMETRIC DEGREES FROM FOVEA

FIG. 1. Distribution of cones and rods on and near the horizontal meridian. (From G. Østerberg, 1935.)

* Paper presented by invitation at the Symposium on Vision at the meeting of the Optical Society of America, New York, New York, March 5–6, 1943.

However, there are other functions in which the transition from the one system to the other is fairly sharp and, therefore, in which the two systems behave much more independently. The two curves contained in Fig. 2 show the relation between the wave-length and the relative energy required to produce a specific visual effect at high and at low intensities, respectively. The lower curve contains the data from Hecht and Williams (1922) and represents the relative energy required to produce a barely perceptible brightness after prolonged dark adaptation. The upper curve contains data from Hyde, Forsythe, and Cady (1918) and shows the relative energy needed to produce a given brightness when the fovea (rich in cones) only is involved. The two curves are drawn accurately from their separate data; the vertical separation, however, has been so arranged that the curves are nearly coincident in the red end of the spectrum. This is a graphic expression of the fact that the colorless (rod) and color (cone) thresholds of the eye are nearly identical in the red end of the spectrum.

Starting at the lowest energy level and moving vertically along any ordinate, it is seen that the light of any specified wave-length produces its first visual effect when its magnitude reaches the curve for the response of the rods. This particular function continues to be controlled by the rods as the energy increases until the curve of cone response is reached. At this level of energy, if the activity of the cones is recognized independently, a definite change of the character of the function should be in evidence and should be maintained as the intensity increases. Two such distinct visual functions are the sensations of light and of form. Furthermore, a significant feature of Fig. 2 is that the rod-cone interval changes with the wave-length of light and is small in the red and large in the blue end of the spectrum. Also, the *sensation of color* is characteristic of cone function. The range of intensity between the thresholds of the rods and of the cones (Fig. 2) is controlled by rod function and appears colorless. The rod-cone or colorless-to-color interval should be small in the red and large in the blue end of the spectrum. This was shown to be the case by Charpentier in 1880 and by Parinaud in 1898. This difference between the thresholds of the rods and cones is

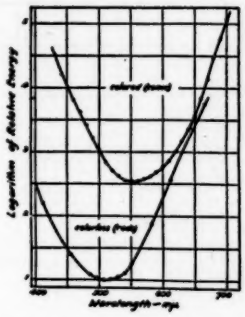


FIG. 2. Relation between wave-length and relative energy required to produce a specific visual effect at high and at low illuminations, respectively. The lower data (rods) are from Hecht and Williams (1922) and represent the relative energy necessary to produce a barely perceptible brightness after prolonged dark adaptation. The upper data (cones) are from Hyde, Forsythe, and Cady (1918) and give the relative energy required to produce a given high brightness, using the fovea (cones) only. The two curves are drawn accurately; the vertical separation has been arranged so that they are nearly coincident at the red end of the spectrum.

referred to as the *photochromatic* interval. As a matter of fact, insofar as intensity and spectral region of light are concerned, all of the visual phenomena to be described and discussed in subsequent paragraphs show a sufficient degree of independence in the functions of the rods and of the cones so that their effects can be distinguished fairly readily and easily understood in terms of Fig. 2.

2. Fundamental Considerations Regarding Dark Adaptation

I. Rods and Cones

The ability of the human eye to increase in sensibility during a period in the dark was described first by Aubert in 1865. Since that time the phenomenon of dark adaptation has been investigated frequently in man and in other animals. Excellent summaries and reviews of the subject have been prepared by Dorothy Adams (1929), Müller (1932), and Hecht (1934). The earliest measurements were made by Piper (1903) and were confirmed by Nagel in 1911. The determinations were made with white light, large measuring fields, and peripheral observation. In general, the results of these investigations showed that there was a continuous decrease of threshold during a period of about

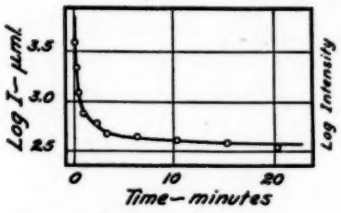


FIG. 3. Dark adaptation of the fovea as measured with red light. Each point is the average of thirty measurements with fifteen subjects. Intensities are in micromillilamberts. (From Hecht, 1921.)

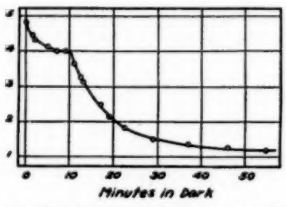


FIG. 4. Dark adaptation of the eye as a whole. Measurements of Engelmann's eye made by Piper (1903). Note the initial stage of cone adaptation followed by rod adaptation.

thirty minutes and covering a range of light intensity of about 5000 units to 1 unit of intensity. The marked change of light sensitivity was ascribed correctly to the rods, since the retinal areas stimulated were in the periphery and there was absence of color sensation at the threshold.

The early investigators (for example, Piper, 1903; Nagel and Schaefer, 1904; Dittler and Koike, 1912) generally accepted the view that the rods only could adapt themselves to the dark and that the cones could not do so, although there was some evidence to indicate that the cones did adapt themselves slightly to very low illumination. In 1921 Hecht definitely settled this question when the dark adaptation of the *rod-free* area of the fovea was measured accurately. The reasons for the previous failures or incorrect hypotheses or beliefs also became apparent. Figure 3 shows the data obtained by Hecht covering the dark adaptation of the fovea as measured with red light. It is apparent that *foveal dark adaptation* is very rapid and normally is consummated in three to five minutes. If both rods and cones show dark adaptation but with different rates and different threshold levels, measurements of the eye as a whole (in areas, for example, 5 to 10 degrees peripherally) should show both rod and cone adaptive phenomena. In 1922 Kohlrausch demonstrated that, in a retinal area containing both rods and cones, there was a definite break in the curve of dark adaptation. Prior to the distinct point of flexion in the curve there is a period of dark adaptation which is a function of the cones and after the break there is a course of dark adaptation which is due to the rods. In his review on rods, cones, and the chemical basis of vision, Hecht wrote:

> It is interesting to record that on re-examination, some of Piper's 1903 measurements, as well as some made later (Piper, 1905), show this break strikingly, but that it was obscured by the absurd method of plotting adaptation then in vogue. For historical reasons, I [that is, Hecht] give in figure 4 [also Fig. 4 in the present paper] the data for Engelmann's eye taken from Piper's 1903 paper. It is apparent that the primary drop in threshold is rapidly over, as is the case with the foveal cone adaptation in figure 3 [also Fig. 3 in the present paper] while the secondary drop in threshold appears later and proceeds for over 30 minutes.

The relative ranges of intensity of illumination (in millilamberts) concerned with cone and rod vision, respectively, are shown in Fig. 5. In the diagram, the change-over between rod vision (low brightness levels) and cone vision (high brightness levels) is indicated at about 0.01 millilambert. Actually, the transition takes place

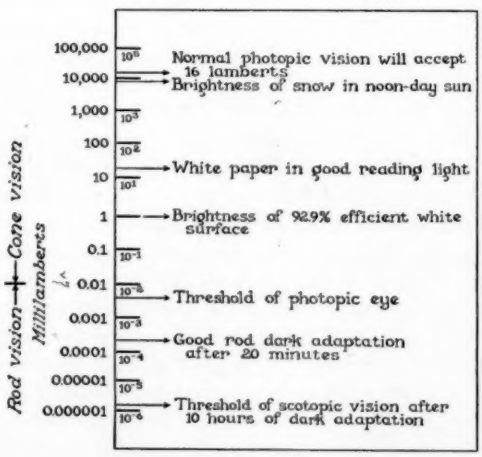


FIG. 5. The relative ranges of intensity of illumination concerned with cone (high brightness levels) and rod (low brightness levels) vision, respectively. The transition between cone and rod vision is shown as though it occurred suddenly at 0.01 millilambert. The transition actually takes place more gradually in the range of 0.1 to 0.01 millilambert.

gradually in the range of 0.1 to 0.01 millilambert. Even in the range between 1.0 and 0.1 millilambert, where cone vision is operative, deterioration of visual acuity and of discrimination of intensity with lowering of the brightness level becomes quite noticeable. The degree of sensitivity which develops in the retina after a rest in the dark is worthy of note and, in the language of the street, is "something to marvel at." After ten or twelve hours in darkness, the threshold brightness reaches a lower limit of 0.000001 millilambert if white light is used. Yet that same retina, after exposure to bright light for a few minutes, enters a state which is referred to as light-adapted, in which its threshold (that is, the smallest amount of light which will produce the sensation of vision or the threshold of photopic vision) has risen to about 0.005 millilambert and in this state the retina is capable of accepting, without injury to itself and with excellent transformation of these energies into cerebral values of vision, energies as high as 16,000 millilamberts. In other words, its threshold has increased to about 5000 times the threshold of the dark-adapted eye and the maximal brightness that is acceptable by the completely light-adapted retina is more than 10,000,000,000 times the threshold of sensitivity of the completely dark-adapted eye (Fig. 5). The most striking thing about the change of visual threshold, as one enters and remains in a dark room, is its range; it can easily cover a gamut from 100,000 units of light intensity at the beginning to 1 unit at the end of dark adaptation.

II. Color and Preadaptation

If the primary and secondary (or initial and final stages of) adaptations shown in Fig. 4 are concerned fundamentally with the functioning of cones and rods, respectively, it should be possible to isolate them by means of their visibility curves. In the extreme red portion of the visible spectrum it is to be noted that the final thresholds of the rods and cones are about the same (Fig. 2). Hence cone adaptation only will be elicited when the course of dark adaptation is investigated by the use of red light. On the other hand, if blue light is used, after the initial rapid cone adaptation, the threshold should

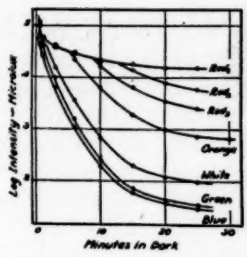


FIG. 6. Curves of dark adaptation obtained with an area 1 degree in diameter, situated 5 degrees above the fovea. Red_1 is the only color which limits the measurements to cones. The other two red filters let through more orange and yellow radiation and hence demonstrate rod adaptation. Cone adaptation is barely perceptible with blue light. (From Kohlrausch, 1922 and 1931.)

continue to decrease because of the low level to which the responses of the rods finally sink. A series of intermediate results would be expected for various colored light between the extreme blue and the extreme red end of the spectrum. In 1922 Kohlrausch put these conclusions to experimental test by investigating the dark adaptation of a small retinal area 5 degrees from the macula, using light stimuli of different colors. A reproduction of Kohlrausch's data is given in Fig. 6. It is evident that, for the majority of colored light stimuli, there are two distinct parts to the process of dark adaptation. The first part corresponds to adaptation of the cones and is nearly the same for all colors which show it, namely, the red, orange, and yellow portions of the spectrum. This cone adaptation, with low intensities of light adaptation and short periods of exposure, is over in about three minutes. This is followed by dark adaptation of the rods. Depending on the photochromatic interval, rod adaptation appears earlier and reaches lower thresholds as the light used for retinal stimulation moves to the blue end of the spectrum.

The measurements of Kohlrausch show little if any cone adaptation when green or blue light is used. The preliminary period of light adaptation was not long enough in exposure or high enough in intensity to bring out cone adaptation. In 1936 Hecht and Haig found that, if the light adaptation is sufficiently intense, there is evidence of cone adaptation. The results with a group of different intensities of retinal light

adaptation are shown in Fig. 7. It is evident, in general, that the more intense the light adaptation the more pronounced and prominent is the cone adaptation and, therefore, the later the appearance of rod adaptation. Furthermore, the difference in the rates of cone and rod dark adaptation is associated with an inverse speed of light adaptation. Apparently the rods adapt to a given light intensity more rapidly than do the cones. At any rate, this is the conclusion of Müller who, in 1931, measured the dark adaptation subsequent to the preadaptation of the retina to white light of about 3000 luxes for different periods. After one minute of light adaptation only the rod dark adaptation was present. Longer exposure to the preadapting light appears to change neither the rate nor the extent of the dark adaptation but acts to delay its appearance. Cone adaptation, under the criteria imposed, was demonstrable after a two-minute period of preadaptation and became more evident the longer the preceding period of light adaptation. The rods apparently reach a photostationary state in the light more quickly than the cones. Wald and Clark in 1936 confirmed the findings of Müller. Subsequently many other careful investigators have obtained corroborative data.

Figure 8 shows the relation between the intensity of adaptation and the time required for the eye to reach a given threshold in the dark. The lower data are for a colorless threshold, indicating rod function, while the upper data are for a colored threshold and indicate cone function.

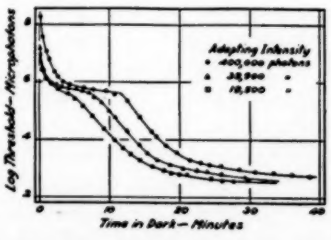


FIG. 7. The curves of dark adaptation as measured with violet light subsequent to different degrees of light adaptation. These curves show that there is evidence of cone adaptation, as measured with violet light, if the intensity of light adaptation is sufficiently high. The filled-in symbols indicate that a violet color was apparent at the threshold, while the unfilled symbols indicate that the threshold was colorless. (From Hecht and Haig, 1936.)

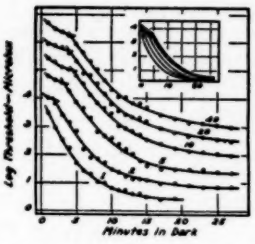


FIG. 8. Dark adaptation of the eye after preadaptation to 3000 luxes for one, two, five, ten, twenty, and forty minutes, respectively. The time axis is the same for all curves; the ordinates are applicable only to the lowest curve; each curve is displaced 0.5 log unit above the preceding curve. The inset at the right hand brings the data together and shows that the longer the preadaptation period (a) the more evident is the dark adaptation of the cones and (b) the more delayed is the secondary rod adaptation. (From Müller, 1931.)

Any two colored lights are effective in some constant energy ratio for the stimulation of the cones and in some other fixed ratio for stimulating rods, because of the different spectral sensitivities of these two types of receptors. Such ratios can be used to analyze precisely the participation of rods and of cones in visual functions. It is possible to measure the absolute thresholds for the sensation of light, regardless of color, alternately through yellow (Wratten 73) and violet (Corning 511) filters. At about 13 degrees from the fixation point (0.75 degree field) the cone threshold for violet lies 0.8 to 1 log unit above the cone threshold for yellow light. As the area of exposure is moved centrally the cone separation ultimately widens 1.2 log units within the fovea, whereas the rod separation correspondingly narrows because of the disproportionate absorption of violet light by macular pigment. Measurements in the completely dark-adapted eye showed that the rods govern the threshold beyond about 2 degrees from the point of fixation. Within the 2-degree area, the cones participate in the threshold levels to an increasing amount and finally exclusively as the fovea only is exposed. Within the fovea a parallel pair of curves for the two colors (yellow and violet) may be obtained, of a pure cone separation throughout. In peripheral fields, after a period of pure cone separation, the curves approach each other, cross, and then rewiden to the pure rod separation. The region of mixed rod and cone function is extensive; at 0.5 to 2.5

degrees eccentrically the major portion of the range of dark adaptation is included. These data and other results of a similar character have been found independently by Wald and by me (data as yet unpublished). These findings definitely indicate that the presence or absence of the sensation of color is not a wholly reliable criterion of cone or rod function. As Wald concluded: "Color sensations are still experienced when the shift from cone to rod vision is well advanced, and cease long before the shift has been completed."

III. Retinal Location

As was pointed out in the introductory paragraphs and as shown in Fig. 1, the histologic structure of the retina varies in different areas according to a definite pattern. The fovea centralis is the central visual area of about 1.5 degrees, which is rod-free, while the slightly larger area, the macula, with a diameter of 2 degrees, contains so few rods that it may be considered as free from rods. In other areas, the retina contains rods and cones. Østerberg (1935) has shown that the number of cones per unit area decreases from the center toward the periphery, quite rapidly at first and then more slowly, whereas the number of rods per unit area gradually increases out to about 18 degrees and then slowly decreases as one proceeds peripherally. For the greater part of the retina the ratio of cones to rods in an area of any specified size decreases as the area is moved outward from the macula. Hence, in terms of histology and on the premises of the duplicity theory, it follows that measurements on dark adaptation, using white light, should show an almost pure cone response when restricted to a 2-degree central area, and a combination of rod and cone behavior for larger areas with central fixation. These deductive conclusions were verified experimentally by Hecht, Haig, and Wald in 1935. Figure 9 shows the data secured by these investigators using white light and a series of centrally fixated fields. With a 2-degree field, the dark adaptation is chiefly of a cone character with some secondary (presumably rod) adaptation in evidence after about fifteen minutes. For areas larger than 3 degrees, rod adaptation is present without question. The thresholds of rod adaptation appear earlier and the adaptation assumes lower levels as the size of the area increases. Beyond 20 degrees there is no appreciable change with increase of area of retinal stimulation.

The cone responses for areas larger than 2 degrees are not the same as for the macular (2 degrees) field. This result may be explained on the basis that, as the area of retina under test is increased, there are both rods and cones outside of the fovea which are responding. A significant statement, made some years ago by Hecht and subscribed to by many investigators in this field, is to the effect that it is not to be expected that the thresholds and rates of adaptation of these peripheral cones should be the same as those of cones in the central area (fovea). And yet, in spite of the truth of this statement, we find many comparisons of responses of cones in the periphery with responses of rods in similar areas, whereas comparisons of such data should be between cones in the macula (2-degree area) and rods in the densely populated peripheral areas (15 to 20 degrees from the center). The chief functions of the cones are distinctness of vision (form sense) and discrimination of color; the chief function of the rods is the response to minimal light stimulus (light sense).

Well-established and authentic data exist which describe the relation between the size of the field and the minimal intensity of light required to make it visible. A satisfactory summary to the date of publication (1914) was given by Parsons. As Hecht stated:

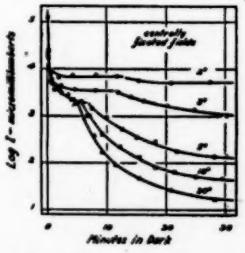


FIG. 9. Curves of dark adaptation for the cones and rods for centrally fixated areas whose diameters (in degrees) are shown in the diagram. The larger the field, the sooner does the secondary rod response appear and the greater is the range of intensities covered. (From Hecht, Haig, and Wald, 1935.)

At first glance, it might seem that the decrease in threshold which shows itself with the larger areas is due to the area-threshold relationship. However, it is simple to show that the major factor involved in these data of dark adaptation and final threshold has almost nothing to do with area but rather with the fact that the retina is not a uniformly sensitive surface. Coupled with its histological structure already referred to, it possesses a permanently graded sensitivity. This may best be illustrated in figure 10 [also Fig. 10 in this paper] which shows the dark adaptation of a 2° field placed in different retinal positions. In the center at 0° off the central axis, adaptation is essentially cone and, as before, the adaptation curve shows the usual rod-cone dichotomy. The final threshold drops still more as the area is moved out to 5° and 10° from the center.

IV. Area and Visual Threshold

In 1938 Wald published his work on area and visual threshold. He was able to show that the simple properties of a mosaic retina account qualitatively and quantitatively for the characteristic variation of the visual threshold with field area. Wald and Haig obtained data on dark adaptation in fields of 1, 2, 3, and 5 degrees in angular diameter, respectively, fixated at 15 degrees above the fovea. Figure 11 gives the correct thresholds for the 5-degree field; the remaining curves have been displaced uniformly on the axis of the logarithm of the threshold (log threshold) in order to emphasize their identity of form. Table I (Wald) contains data regarding number of measurements, area, average log threshold, and computed log threshold. These plotted results and data show that the shape of the dark adaptation curves remains unaltered as the diameter of the field is increased from 1 degree to 5 degrees. Apparently, within these limits the elementary composition of the fields remains constant. However, in these homogeneous regions of the retina, pronounced effects of area on the threshold levels are found. At 15 degrees above the fovea, the threshold for a field of 5 degrees diameter is only a seventh of the threshold for a field of 1 degree diameter; at 25 degrees above the fovea, the reduction of the value of the light threshold is to a tenth.

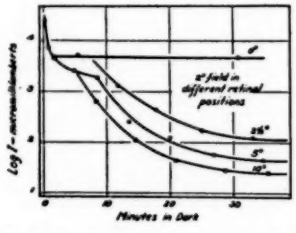


FIG. 10. Dark adaptation as measured with a 2-degree retinal stimulus area placed at 2.5, 5, and 10 degrees from the center (0 degrees). Comparison of these data should be made with the data in Fig. 9 for centrally fixated fields of different size. It is to be noted that the farther in the periphery the measurements are made, the sooner does the rod adaptation appear and the lower is the final threshold. (From Hecht, Haig, and Wald, 1935.)

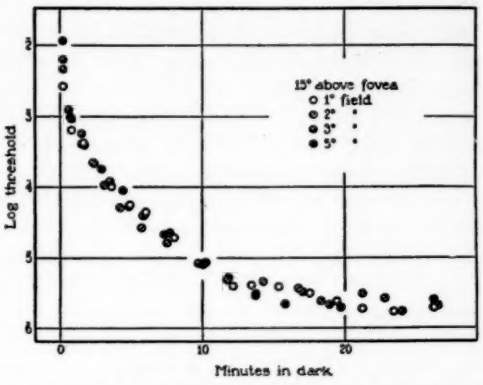


FIG. 11. Dark adaptation in fields fixated 15 degrees above the fovea. The thresholds for the 5-degree field are in millilamberts. The remaining curves have been displaced uniformly on the "log threshold" axis to emphasize the identity of form. (From Wald, 1938.)

Wald made a very critical analysis of the experimental data and showed that, ultimately, only the basic concept of the retina as a popula-

TABLE I. Data concerning the effects of size and location of stimulating fields on the thresholds. (From Wald, 1938.)

Number of measurements	Area (sq. cm)	Average log threshold (millilamberts)	Computed log threshold (millilamberts)
	15° above fovea		
9	0.301	$\bar{6}.82$	$\bar{6}.81$
9	1.258	$\bar{6}.24$	$\bar{6}.28$
12	2.832	$\bar{6}.14$	$\bar{6}.14$
9	5.216	$\bar{6}.06$	$\bar{6}.05$
9	8.696	7.97	7.97
	25° above fovea		
6	0.301	$\bar{6}.99$	$\bar{6}.98$
8	1.258	$\bar{6}.43$	$\bar{6}.43$
10	2.832	$\bar{6}.16$	$\bar{6}.21$
7	5.216	$\bar{6}.03$	$\bar{6}.06$
7	8.696	7.98	7.94

In fields fixated 15° to 25° above the fovea, the shape of the dark adaptation curve remains unaltered as the field diameter is increased, indicating that the elementary composition of the fields remains constant.

tion of relatively independent units is indispensable to the analysis. If this is accepted, the characteristic type of dependence of all retinal properties upon area follows inevitably. Specifically applied to the area-threshold relationship, these considerations reduce to two propositions: the first implicit in the homogeneity of the retinal areas examined, the second a reasonable assumption concerning the threshold. The very logical assumption may be made that the threshold corresponds to the stimulation of a constant number of elements. Though any type of function for the distribution of thresholds yields the correct qualitative area-threshold relation, a "true" distribution is needed for a quantitative description of the phenomenon.

Through the assumption of the constant threshold number, the requisite portions of this distribution may be derived directly from the data. If one desires the threshold distribution in the 1-degree field 25 degrees above the fovea (see the data of Table I) it may be found by the following procedure. At log $I=\bar{6}.99$, the constant threshold number of elements n_t is active in this field of *relative* area 1.00. At log $I=\bar{6}.43$, n_t elements operate in the 2-degree field of relative area 4.18; therefore, at this intensity $n_t/4.18$ elements are active in the 1-degree field. At log $I=\bar{6}.16$, n_t elements are stimulated in the 3-degree field of relative area 9.42; hence, at the same intensity $n_t/9.42$ elements are active in the 1-degree field. By completing this process five points (since five areas are specified in Table I) are obtained on the distribution curve which correlates the illumination to the relative number of active elements in the 1-degree field. The distributions in larger fields are simple multiples of this, proportional to area. The points obtained in this manner for a 1-degree field, 25 degrees above the fovea, are plotted in Fig. 12. The points themselves adequately complete a quantitative analysis. This may be recapitulated: 1. Using the assumption of a constant threshold number, the relevant portion of the threshold distribution in one area is obtained from the area-threshold data. 2. The distributions for other areas are simple multiples of this. 3. The intercepts of these distributions with the constant threshold number at n_t regenerate the original data.

A general equation for the type of distribution outlined in the foregoing paragraph is useful since a rational formula for area-threshold relations thus may be made possible. Obviously the distribution function must end by becoming parallel to the abscissa, when the total number of elements in the area becomes active. The complete function is, therefore, *sigmoid* in form when threshold is plotted logarithmically. A number of formulas for sigmoid curves may be applied; any one of them yields roughly the right type of answer. If one takes such a naïve distribution as would be obtained by a linear relationship between the number of elements n, and the retinal property x (x=intensity of illumination), then the equation of the lines would be

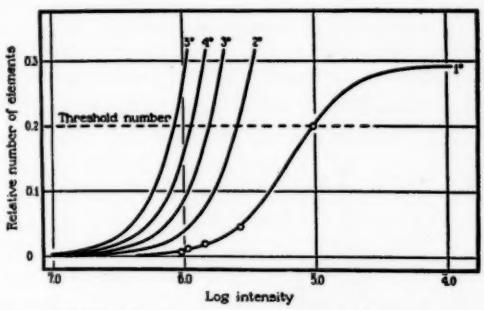


FIG. 12. Graphic analysis of the threshold-area relationship, 25 degrees above the fovea. The open circles were calculated from the averaged data of Table I as discussed in the text. The distribution curves conform to the equation $0.54 \log (A/n-1)+\log I=-5.20$. The line of constant threshold number cuts them in theoretical thresholds for the various areas. (From Wald, 1938.)

$n=KAI$ in which n is the number of active elements; A is area; $I=x$=intensity, and K is a proportionality constant. If n is held constant to the threshold value n_t, the equation becomes $AI=n_t/K=C$, a constant. This relation is the familiar *Riccò's law* (1877). As Wald stated: "It is inexact but encouragingly close to a correct solution."

There has been one expression which has been very successful in describing the data, namely,

$$(A-n_t)^k I=C,$$

in which A=area; n_t=constant threshold number of elements; I=threshold intensity; and k and C are constants. This general type of equation may be derived from either of two sources,

the first of which is Hecht's equation

$$KI = x^p/(a-x)^q,$$

in which I = intensity, x = concentration of photoproducts, a = maximal value of x, and K, p, and q are constants. Rewritten in the form of threshold distribution, this equation becomes

$$KI = n^p/(A-n)^q$$

or

$$(A-n)^q I = n^p/K = \text{constant}.$$

The second derivation does not assume any mechanism. It is based on Verhulst's so-called logistic formula (1838), a symmetric sigmoid function. This is of the form

$$y = \frac{K}{1+Ce^{-mx}},$$

in which y and x are variables, K is the maximal value of y, and C and m are constants. As a threshold distribution this may be written

$$n = \frac{A}{1+Ce^{-m \log I}},$$

or

$$nCe^{-m \log I} = A-n.$$

When n is held constant to the threshold value n_i

$$k \log (A-n_i) + \log I = k \log n_i C = \text{constant},$$

or, removing the logarithmic notation,

$$(A-n_i)^k I = \text{constant}.$$

Such types of analysis lead to the general expression $(A-n_i)^k I = \text{constant}$. When the threshold number of elements n_i is a small part of the total number in the visual field A, the term $(A-n_i)$ approaches A as its limiting value. This condition exists in quite small foveal fields, in which the concentration of receptor units (individual cones) is high. It should obtain also in much larger peripheral fields, since the retinal elements in the periphery are comparatively large summating groups of rods. In both instances the threshold-area equation reduces to the simple form $A^k I = \text{constant}$. Interestingly enough, Abney first empirically discovered this expression. If, with n_i a small fraction of A, k chances to take the value 0.5, the equation then becomes $\sqrt{A} \cdot I = \text{constant}$, or ***Piper's rule.***

A number of investigators have measured the area-threshold relation in small fields centrally fixated. If the diameter of the fields does not exceed 1.5 degrees, they may be assumed to fall wholly within the fovea and to stimulate a relatively homogeneous population of cones. The results of the experiments of Abney in 1897 and Piéron in 1920 yield straight lines in the case of a direct plot of the log area against log threshold. Such a relationship was discovered empirically by Abney. Apparently the threshold number of elements n_i is a negligible fraction of the total populations of these fields.

Riccò (1877) and Charpentier (1882) proposed the empirical rule, AI = constant, for foveal fields. This expression states that the liminal foveal response is evoked by a constant flux of light regardless of its spatial dispersion. Many demonstrations and proofs that this simple relationship is not true have been furnished (for example, Abney and Watson, 1916, and Piéron, 1920) but apparently its simplicity has preserved it.

V. Night Blindness

Night blindness and similar visual disturbances have been associated for years with the nutritional state of the body. In recent years this connection has been traced to the vitamin A of the diet and has become explicable in terms of the chemical relation between vitamin A and the light-sensitive pigments of the retina. Hemeralopia is a condition in which vision at low intensities of light is decidedly poorer than that of normal persons. It generally becomes manifest to its possessor by an inability to see objects or to move about with certainty in dim lights at night. This condition of poor night vision was noted centuries ago. From 1881 its association with deficient rod vision has been recognized (Parinaud, 1881; Treitel, 1885).

There are two distinct varieties of night blindness. One of them is relatively temporary; it is caused by vitamin A deficiency, as was demonstrated by Holm (1925), Fridericia and Holm (1925), and Tansley (1931). They demonstrated that animals (that is, rats in general) become night-blind when fed for a few weeks on a diet deficient in vitamin A. The dietary type of

vitamin A deficiency is understandable in terms of the connection between vitamin A and visual purple demonstrated by Wald in 1936. The other variety of poor night vision is inherited (known as Oguchi's disease in Japan) and is apparently incurable. The suggestion has been offered that probably some other constituent of rod function is missing in this condition.

Subsequent to Piper's first measurements on dark adaptation and their association with rod function, it was an obvious procedure to test the dark adaptation of night-blind persons. It appears that Heinrichsdorff first made such a test in 1905 with Piper's apparatus. Since the proper conditions for making such tests, particularly the necessity of proper preadaptation to light, were not understood fully at that time, it

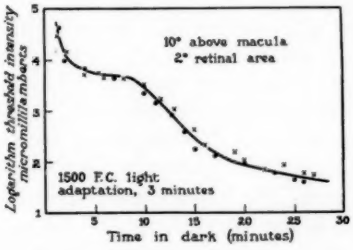


FIG. 13. Curve illustrating the two portions of dark adaptation; one portion rapid (cones), the other (rods) slower in recovery. The black dots (●) and crosses (×) represent data obtained under similar conditions, time of test and ingestion of food on two different occasions. (From Steffens, Bair, and Sheard, 1940.)

remained for Messmer (1908) to show clearly a diminished dark adaptation in hemeralopes and, in some cases, the complete absence of dark adaptation. Much later and apparently independently Takagi and Kawakami in 1924 measured the dark adaptation of several persons who had hereditary night blindness (presumably retinitis pigmentosa) and found that, for the first two hours in the dark, such persons showed practically no change in threshold, while under the same conditions of measurement the threshold level of the normal eye had dropped to about 1/5000 of its initial value in thirty minutes. Rod adaptation apparently is lost almost completely in such pathologic conditions, or at least its speed is greatly retarded and its extent much reduced. In 1929 Dieter confirmed these findings. From these investigations it is apparent that, for the first hour at least, dark adaptation of hemeralopes shows only the primary drop in the threshold of the cones and no secondary rod drop. The evidence of Dieter is particularly conclusive because he found, under all conditions of brightness, that his hemeralopes possessed only one visibility curve, which corresponded to his own high intensity cone-visibility curve. This natural experiment, therefore, bears out the other data and finds its explanation in terms of the independent functioning of the rods and cones.

3. Criteria for Precise Measurements of Dark Adaptation

In brief review of information which has been given in some detail in the foregoing paragraphs, let it be stated categorically that there are at least eight specifications which must be made in order that measurements of dark adaptation may be numerically precise and quantitatively valid. These conditions are:

1. Accuracy, constancy, and reproducibility of physical measurements and of various portions of the ensembles.
2. The intensity of the light used for light adaptation.
3. The duration of the exposure to light before starting measurement on the course of dark adaptation.
4. The size of the area of retinal test.
5. The retinal location.
6. The color of the light used for the determination of threshold levels.
7. The duration of the exposure of the retina to the light used during the course of dark adaptation (intermittent exposure).
8. The size of the pupil.

Figure 13 (Steffens, Bair, and Sheard, 1940) again emphasizes the fact that dark adaptation proceeds in two steps. The first step is rapid and over in a few minutes; the second is late in starting and proceeds for a half-hour or more. These two portions are records of the functions of the cones and rods, respectively. When the measurements have been made with violet light this fact is brought out by the sharp and quite readily noted color difference between the thresholds which are determined by the cones and by

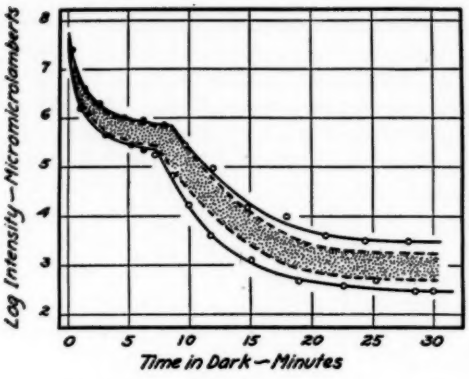


FIG. 14. The course of dark adaptation in a group of subjects. The upper and lower curves are of the two persons who yielded the highest and lowest final rod thresholds, respectively, of the group of 110 persons tested. The filled-in circles (●) represent colored response and the open circles (○) indicate colorless light vision. The stippled area contains 80 percent of those tested. (From Hecht and Mandelbaum, 1939.)

the rods. Even an inexperienced observer records the fact that, at the threshold, the light observed in the first portion (see the solid circles in Fig. 14) appears violet or blue, while the light observed in the second section (unfilled circles) does not show any color at the threshold.

The eight conditions under which the measurements are made greatly influence the relative extent, the duration, and even the form of the two parts of the dark adaptation curve. Thus, the brightness and duration of the light adaptation will determine the time of the appearance of the cone-rod transition. After prolonged exposure to high intensities of illumination, the break may not occur before about fifteen minutes, whereas after short exposure to low intensities the transition may come after a minute or even less. The same is true for the color of the light used for obtaining the course of dark adaptation. If dark adaptation is measured with red light there will be a dominant cone section with a very slight rod response, which may appear after twenty to thirty minutes; if violet light is used, the cone portion may scarcely show and only the rod portion will be present. In the same way the size of the area of retinal stimulation and its location will share with the other factors in the control of the course of dark adaptation. Furthermore, the size of the pupil will exert its effect upon the values of the thresholds of dark adaptation and should be measured and mathematically considered in the determination of the values of dark adaptation levels. Likewise, the consideration of the size of the pupil is of importance (as has been emphasized by Tansley and other investigators) in the determination of normal standards and in the comparison of data in clinical cases and in certain meticulous tests.

However, in the case of measurements of pilots and night fliers and in certain types of aeromedical investigation such as are now being conducted in a few institutions under the war emergency program, I am of the opinion that measurements on dark adaptation should be taken with both eyes of the subject exposed (having no regard for ocular dominance or relationships of accommodation and convergence, binocular single vision, and other factors) and with the pupils as they are, irrespective of their size or photostatic activity, for the reason that the data should be obtained on the pilot under his own unhampered responses to the environmental conditions that are imposed. These environmental conditions should simulate actual flying conditions as closely as possible.

4. Standard Conditions, Apparatus, and Methods

Many investigators, following the technique of Hecht and his associates, and perhaps using the adaptometer of Hecht and Shlaer, have chosen a light adaptation of sufficient intensity and duration that both cone dark adaptation and rod adaptation are separately and adequately shown in the measurements. This requires a light adapting field, occupying about 40 degrees visual angle, with a brightness of about 1500 millilamberts. The exposure is usually three minutes. The test field occupies 3 degrees visual angle in diameter; this is done on the basis that it is a compromise between a smaller field, which reports the behavior of a uniform part of the retina, and a larger field, on the other hand, which is seen more easily by untrained subjects. Several investigators have used a test field which was viewed 7 degrees nasally with the right eye. This position has been selected for the reason that, at this location, the

population of retinal rods is more nearly equal to that of the cones (although still not comparable in value) than in the center of the retina (chiefly cones) or farther out in the periphery (largely rods). Furthermore, the luminous fixation point (preferably dark red) used to maintain the eye well centered and steady is sufficiently remote from the retinal area under test with regard to its function of dark adaptation so as not to interfere with this function.

The light used for retinal stimulation and for the determination of the threshold of dark adaptation is frequently violet. The use of the extreme violet end of the spectrum makes the rod response maximal in range and enables the observer to make clear distinction between cone function (color) and rod function (colorless) to which reference has been made in preceding paragraphs. The light used for measurement of dark adaptation thresholds is exposed in flashes of one-fifth second whenever intermittent retinal stimulation is employed. This period of exposure is long enough to produce a good perception of the test area and brief enough to be close to the retinal action time. These specifications and desirable procedures may be fulfilled by an adequate control of intensities of light, periods of time, positions or retinal locations, and areas.

The fulfillment of these essential specifications and procedures may be accomplished by the use of an optical bench carrying the various accessories. Such an ensemble is diagramed in Fig. 15. With this arrangement of equipment and following my technique and procedures, the subject's right eye (the left eye being occluded) is light-adapted for three minutes by gazing upon a white screen with a uniform brightness of about 200 millilamberts. This is a very low light adaptation as compared with the intensities (1500 millilamberts) used by many other research workers. It was selected for the reasons that (1) the maximal dark adaptation of the rods is obtained in a minimum of time, (2) the adaptation functions of rods and cones are investigated separately by the use of small test fields (ordinarily $\frac{1}{3}$ to $\frac{1}{2}$ degree visual angle in diameter) at the macula and at 10 degrees paramacularly, and (3) exposure to very bright sources of light often produces headaches (doubtless photophobic in character) and other untoward symptoms among patients who are undergoing clinical tests. It has been our custom to dilate the pupil of the eye under test with a 1 percent solution of euphthalmine hydrochloride. The diameter of the pupil is measured with the Moss modification of the Broca pupillometer. All calculations of the threshold levels (reduced to a standard pupillary diameter of 5 mm) are made according to the simple formula

$$I = K(I_1 FP/D^2),$$

in which I_1 is the brightness of the target (retinal-stimulus target A, Fig. 15) in micromillilamberts at a specified distance D_1 of the light source (E, Fig. 15) from the target, F the value of the light transmission of the neutral filter

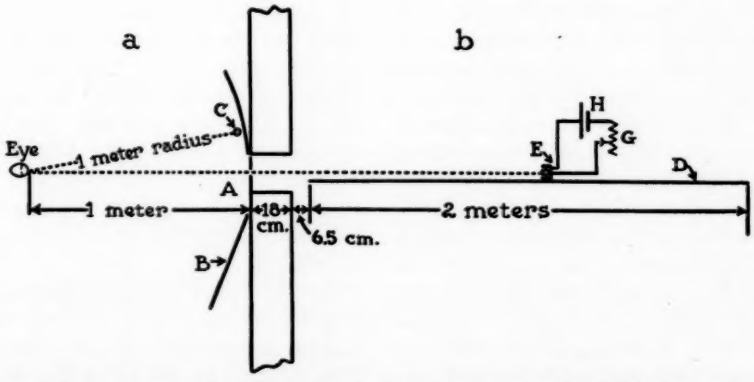


FIG. 15. Diagrammatic sketch of optical bench and accessories which serve as a satisfactory dark adaptometer. (From Steffens, Bair, and Sheard, 1940.)

used, P the pupillary diameter (mm), and D the position of the light E on the optical bench.

Furthermore, it has been my experience that the data are as accurate and as useful in the solution of various visual problems or in clinical investigations of possible A avitaminosis as judged by measurements on dark adaptation, if (1) white light is used and (2) intermittency or flashing of light is omitted. The preponderance of opinion on the part of children, airplane pilots, and clinical subjects has been favorable to the use of very small fields and no flashing or intermittency of the measuring light. However, one wisely modifies the experimental criteria to suit the problem in hand.

Because of the stimulation to clinical investigation on vitamin A deficiency caused by the work of Jeans and Zentmire in 1934 to 1936 using dark adaptation as a criterion and because some of these investigations and other publications have been found to be perhaps more enthusiastic than critical, it is of interest to note that some of the justifiable criticism has centered around the so-called biophotometer, its physical calibration and constancy, and the method of procedure with this instrument. Steffens, Bair, and I, in the course of some investigations on dark adaptation and dietary deficiency in vitamin A, made use of the biophotometer in measuring the thresholds of three subjects over a period of thirty days and compared the results obtained simultaneously with our own experimental ensemble. However, the manner of making determinations of threshold levels was modified from that described by Jeans. Instead of using both eyes, the left eye was occluded and the pupil of the right eye was dilated with euphthalmine hydrochloride. The left upper spot of the quincunx was used as the fixation target and a signal was given as soon as the observer saw the central spot or area of the quincunx. Various sets of data thus obtained showed that the values of the threshold and the courses of dark adaptation were comparable to, and in many instances identical with, the values obtained by our experimental ensemble. Hence it follows that divergent and somewhat conflicting results may be obtained by inaccurate instrumentation on the one hand. On the other hand, through the simultaneous use of various types of visual adaptometers, it may be shown that differences in instrumentation and essentials of experimental procedure are not solely responsible for the variance of findings and conclusions of various investigators.

Without doubt one of the most accurate and satisfactory instruments is the adaptometer of Hecht and Shlaer. Not only does this instrument incorporate all of the essential specifications which have been listed previously but it is so arranged that variations can be made in case it is necessary to investigate other aspects of dark adaptation under different but controlled conditions. The procedure involves light adaptation to 1500 millilamberts for three minutes and measurements, during the ensuing period of darkness, of the threshold of a retinal area 3 degrees in diameter situated 7 degrees nasally for flashes of violet light of one-fifth second in duration. Measurements of dark adaptation made in this manner reach a final threshold whose brightness is about 0.000000001 lambert. This brightness corresponds approximately to the appearance of a white card illuminated by a candle 1000 feet (300 meters) away and corresponds, in turn, to an illumination of 0.000001 footcandle.

In 1941 Wald described his simple portable visual adaptometer, which was built originally for a survey on vitamin A deficiency in northern Newfoundland and Labrador. The instrument was tested and tried under particularly rigorous conditions, since considerable transportation was entailed, often by dog sled and on ski. The ensemble combines precision with maximal simplicity, inexpensiveness, ruggedness, and portability. The usual list of eyepiece and diaphragms, fixation lamp, shutter, and spectral filters is used. The test and fixation lamps are connected in parallel; both are in series with a calibrated 40-ohm rheostat with scale. This arrangement permits the brightnesses of the test and fixation lights to rise and fall together; hence the fixation light is kept automatically at a level appropriate to each stage of visual adaptation.

Many types of visual adaptometers, from the simplest possible to the more accurate and complicated in structure and use, have been brought forth during the period of the war emergency because of the importance of night vision in

aviation. I suspect that many of these instruments, of which I have one or two to contribute, will find their way on to the market, especially if the wave of vitamin victimization carries over into the days when, perhaps, ploughshares no longer will be beaten into cannon.

5. Normal Variation of Findings on Dark Adaptation

Before proceeding to test the effects of dietary deficiencies on dark adaptation or to presume to consider certain experimental data as evidence of either subclinical or frank manifestations of vitamin A deficiency, it is necessary to ascertain the normal distribution of the characteristics of dark adaptation. Hecht and Mandelbaum, following Hecht's own criteria and experimental procedures as previously rehearsed, made routine studies on 110 persons, most of whom had university associations as faculty, students, wives, or children. The reasons given for this choice of subjects are twofold: one of convenience, the other of normality on the basis that university people are more likely than others to be well fed and well balanced in their dietary habits. Much of this is to be questioned, for I have found that teachers and nuns in general are likely to be food faddists and to demonstrate fairly high (hence abnormal) thresholds of dark adaptation. Furthermore, the range of values of the final thresholds of these groups is much greater than other investigators, including myself, have found when young inductees into the armed forces have been tested.

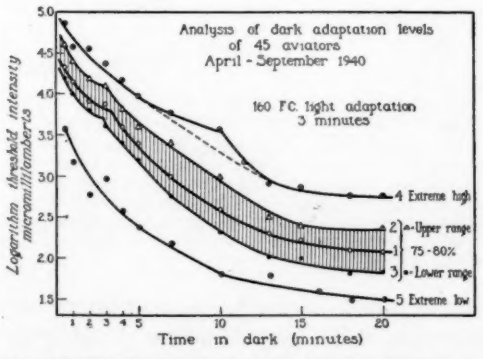


FIG. 16. Distribution of dark adaptation levels of a group of air pilots. The area between curves 2 and 3 includes 75 to 80 percent of all the readings obtained on the group (Sheard).

Be this as it may, Hecht and Mandelbaum's data are shown in Fig. 14; the measurements are all single readings and first performances. The two sets of measurements shown in the diagram (Fig. 14) are of the two persons who yielded the extreme final rod thresholds, the highest and the lowest, respectively. The stippled band between the two dashed curves includes the data on 80 percent of those tested. Hence the chances are high that an average person when tested will give a dark adaptation curve which falls in this 80 percent band. As a matter of fact, statistical studies based on careful tests, adequate controls, and correct standards indicate that not more than 2 percent of the population of the United States show abnormally high or pathologic levels of dark adaptation. However, the subclinical conditions of dietary vitamin deficiency are much more frequent than frank deficiency and are worthy of much further clinical investigation. All of this, however, does not furnish any excuse for everything from corn medicine to cough drops being vitaminized indiscriminately in these United States of America. In other parts of the world and in the care of the military forces, some inclusion of vitamins as accessory foods and protective agencies is highly desirable.

During the past few years I have made several analyses, by various methods, of the data obtained on groups of aviation pilots, school children, and inductees into military service. The methods and equipment have been those described previously in connection with my own work. Figure 16 is an analysis of the dark adaptation levels of a group of forty-five air pilots, ranging from twenty to forty-four years of age, in one of the commercial air lines operating in and out of the city in which I live. The five curves of the plot do not represent, as might be surmised, the best (curve 5) and poorest (curve 4) courses of dark adaptation, with the mean as given in curve 1. All of the data obtained on all subjects tested have been included in the distribution of threshold values for any specified minute during the course of dark adaptation. The band (indicated by vertical lines) between curves 2 and 3 is drawn to include 75 to 80 percent of all of the findings obtained on the group. Hence no single curve represents the course of

dark adaptation of any one person; rather each curve represents the record of the thresholds of some person in the group for the particular range of value (for example, extreme high or extreme low) for any given time during the course of

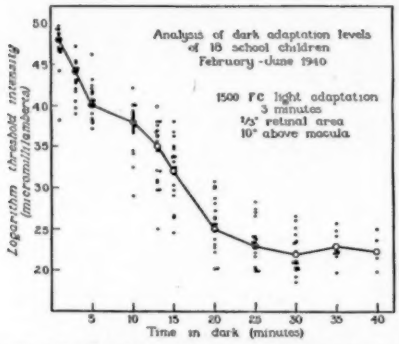


FIG. 17. Distribution and mean of dark adaptation levels of a group of young school children after a preadaptation exposure for three minutes to a field brightness of 1500 millilamberts (Sheard).

dark adaptation. These data and the findings of Hecht (Fig. 14) indicate a range of 0.5 to 0.7 log unit as a possible normal variation. An analysis of the dark adaptation levels of ninety school children of Rochester, Minnesota, between the ages of six and thirteen years, made during the winter of 1940, warrants similar conclusions. The distribution of the dark adaptation levels and the mean curve of a group of eighteen school children are shown in Fig. 17 under the condition of a preadaptation exposure for three minutes to a field brightness of 1500 millilamberts.

In connection with his investigations on the deficiency of inhabitants of northern Newfoundland and Labrador, Wald investigated day-to-day variability. Figure 18 shows the results of tests with binocular fixation; the data are contained within approximately parallel curves, 0.13 log unit wide for the cones and 0.17 log unit wide for the rods. The upper curve with open circles gives the results of one monocular experiment displaced 1 log unit upward. The lower curves with solid circles show the results of four binocular experiments on different days. Thresholds are in micromillilamberts; the field is 2 degrees in diameter and is centered 6 degrees below the fixation point with one-fiftieth second test flashes.

By means of adequate control of a well-trained subject, as well as of environment and procedure, I have been able to repeat dark adaptation thresholds on the same subject from day to day to within 0.1 log unit. This result has been possible even under conditions that might be considered unfavorable, such as the wearing of oxygen masks in low pressure chambers at ground levels or at equivalent higher elevations. In some recent investigations, not yet published, that my associates and I have made it was found that the breathing of oxygen at ground levels (1000 feet [300 meters] in our investigations) produced the best final cone and rod levels and that these levels did not vary more than 0.1 log unit for the same subject on different days. Frequently the variation was as little as 0.05 log unit. However, the general run-of-mine data, with due care, will scatter ordinarily within a range of 0.15 log unit at each sitting and within 0.3 log unit from day to day.

It may happen that for several days or even for a few weeks of measurement of a visual function the person under test is consistent to within less than 0.1 log unit. Then, perhaps, after a short lapse of time or quite quickly as the result of fatigue or of a night out or for no apparent

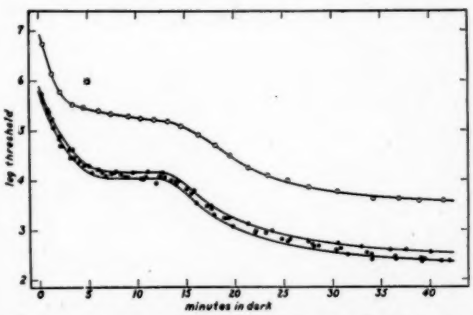


FIG. 18. Dark adaptation following four minutes' exposure to approximately 3000 millilamberts. Open circles: data of monocular test displaced 1 log unit upward. Solid circles: data of four binocular tests on different days. Field 2 degrees in diameter, centered 6 degrees below the fixation point, one-fiftieth second test flashes. (From Wald, 1941.)

reason at all (possibly just being "fed up" with the stress, strain, and monotony of test or even of "life," especially when the element of anoxia has entered into the work) the subject may change and give values which fall 0.3 to 0.5 log

unit above the previous readings. Parenthetically it may be said that there is no reason to believe that a decrease of retinal sensitivity of 0.5 log unit is due, under such circumstances, to a deficiency of vitamin A, since dietary regimen is ample in all particulars. Hence the unfavorable condition of dark adaptation is probably not pigmentary but neural (cerebral) in origin. A range of 0.3 log unit is a factor of 2 in actual light intensity; a range of 0.5 log unit corresponds to a little more than a threefold change. This means that, if a threshold is 1 today, next week or sometime later it may be 2, and usually the values will lie between those limits. This is certainly a small variation when one considers that the total change in threshold during dark adaptation is about 4 log units, or 10,000:1.

Yudkin, making use of the method and instrument devised by him and reported in 1941, found that the final rod threshold varied in the same subject by 0.2 log unit from day to day. The cone threshold varied more, namely, 0.3 log unit. These variations were attributed, not to fatigue, but to related changes in the blood sugar (McFarland and Forbes, 1940).

The results of Hecht, and of Wald, as well as my results, again emphasize that, with sufficiently high levels of light preadaptation, there

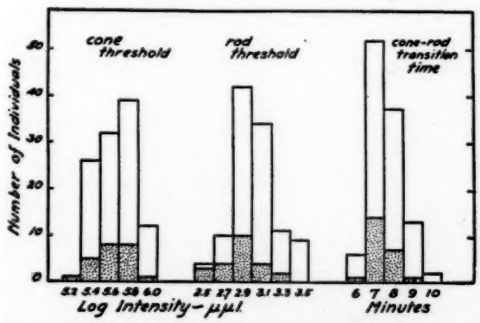


FIG. 19. Distribution of cone threshold, rod threshold, and cone-rod transition time. The stippled areas represent the female portion of the population tested. (From Hecht and Mandelbaum, 1939.)

is exhibited an initial fall of dark adaptation threshold, due to cone function, which reaches a minimal value that is maintained for about ten minutes. The second fall of threshold to a final level in twenty-five to forty minutes (or less, depending on the time of exposure and the brightness of the light adapting field) is due to the rods.

The first published data which I have seen concerning the final cone threshold, the final rod threshold, and the cone-rod transition in the

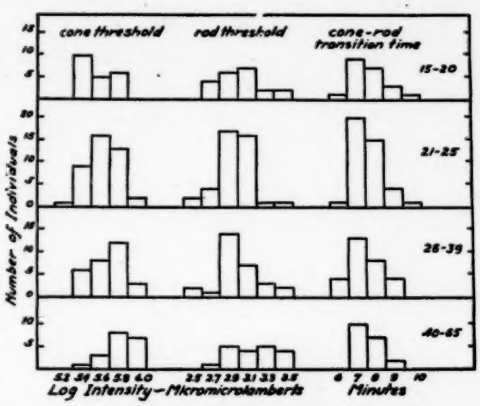


FIG. 20. Distribution of dark adaptation characteristics according to age. The cone threshold is most affected by age, the rod threshold only slightly, and the cone-rod transition time not at all. (From Hecht and Mandelbaum, 1939.)

population tested, are those of Hecht and Mandelbaum. The results are portrayed in Fig. 19. The distribution of cone thresholds appears slightly skewed; the other two distributions are approximately symmetrical. The stippled areas represent female subjects and the clear areas male subjects. Obviously sex does not affect the distribution.

All of us who are concerned seriously with matters of visual adaptation have been cognizant that *age* has some influence on dark adaptation. The cone threshold slowly rises with age, as is shown in Fig. 20. The diagrams showing the distribution of rod thresholds with various age groups show that, from fifteen to twenty years of age, skew is to the left, whereas in the other age groups (twenty-one to twenty-five, twenty-six to thirty-nine, and forty to sixty-five years) skew is definitely to the right of the mode and, in the forty to sixty-five year group, strongly to the right. All of this indicates what some of us have found out from experience, namely, that the youngsters, who are the neophytes in aviation and are less than twenty years of age, as a group have better dark adaptation levels (both for rod and cone functions in general) than the

experienced pilot of the late thirties and early forties. In 1940 Lewis and Haig reported that the range of threshold values obtained for twenty-four children (aged six to twelve years) was about the same as that noted for fifty-four normal adults examined with the same apparatus (Hecht-Shlaer adaptometer) under similar conditions. A slightly higher incidence of 1.6 log brightness (micromicrolamberts) was obtained for children as compared with adults. The mean value and the mode were practically the same for children as for adults.

6. Relation Between Final Rod Threshold and Final Cone Threshold

To anticipate somewhat the presentation of data and facts regarding dietary deficiency of vitamin A, it was discovered some years ago that, for a proper understanding of the chemistry of vision, one significant consideration is the almost parallel behavior of rod and cone thresholds during the periods of deficiency and those of supplementary adequate constituents (the recovery periods) of the diet. This fact must mean that, just as vitamin A enters into the chemical cycle of rod vision because of its association with visual purple, so it also enters into the chemical cycle of cone vision and that the cone sensitive substance (Wald, 1937; Chase, 1938)—iodopsin or visual violet—is probably also a conjugated carotenoid protein like rhodopsin and porphyropsin. There is a visual photochemical cycle involving one or another of the three known photosensitive substances, which are in dynamic relation with their photoproducts (protein, retinene, and vitamin A). The photoproducts, in turn, are the precursors from which the photosensitive substances are formed again. Wald has summarized the direct chemical information now available in the following scheme:

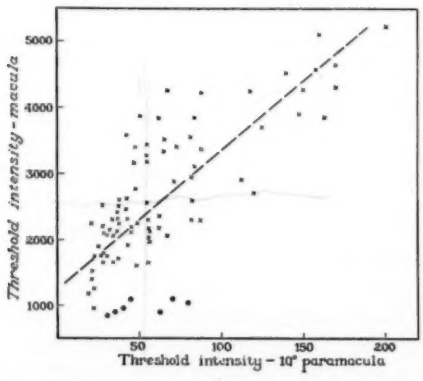


FIG. 21. Relation between final rod threshold and final cone threshold in a group of twenty-four persons. There is a general tendency for the two thresholds to vary in the same direction (Sheard).

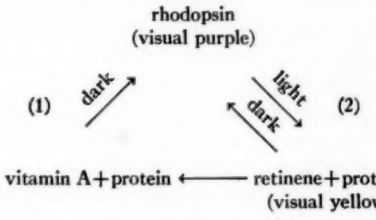


Figure 21 records the final thresholds for the cone and the rod adaptation of a group of twenty-four male high school and junior college students ranging in age from seventeen to nineteen years. All data were taken in the afternoon and about three hours after the noonday luncheon. Insofar as such is possible in such investigations, all conditions were kept constant. Several determinations of final cone and rod adaptation were made on different days on some of the subjects. All of the data were obtained by the method that I have followed, namely, by a direct determination of cone adaptation in the fovea through fixation of a small red light with a field area of 0.33-degree diameter located 1.5 degrees from the fixation light, followed by a determination of rod adaptation using the same retinal stimulus area situated 10 degrees paramacularly. It is evident from an inspection of Fig. 21 that there is a general tendency for the two thresholds to vary in the same direction but that the correlation is none too high. Employing the usual statistical procedures, the correlation coefficient of the two final thresholds (cone and rod) is +0.63, where +1 is a perfect correlation. Hecht made a similar set of determinations on the final rod and cone thresholds (using his own method and therefore finding cone and rod responses in a retinal area 3 degrees in diameter situated 7 degrees nasally) in the population which he studied. He found a correlation coefficient of +0.44.

7. Survey of Threshold Levels of Cones and Rods from the Fovea to 40 Degrees above the Macula

The fovea centralis is the central visual area which is concerned with acuity of form. It is about 1 to 1.5 degrees in diameter, is filled with cones, and is rod-free. The slightly larger area, the macula, with a diameter of 2 to 2.5 degrees, contains so few rods that it may be considered quite free from rods. As the area is increased from 3 to 6 or more degrees, the number of cones per unit area decreases from the center toward the paramacular regions in which the population of rods increases and reaches a maximal value at about 20 degrees peripherally. As Wald significantly pointed out, "at 0.5 to 2.5° eccentrically there is included the major portion of the range of dark adaptation." As will be pointed out in the succeeding paragraphs, there is much evidence that the photochemical processes and pigments for both cones and rods have much in common or at least that vitamin A also enters into the chemical cycle of cone vision and that the sensitive substance, or perhaps one of the cone sensitive substances, is very likely also a conjugated carotenoid protein. Furthermore, there is some evidence that, while cones are cones, not all of them (especially in such a region as the fovea when compared with a region 20 degrees peripherally) may function as acuity organs with the elements supplied with visual yellow.

Again, there is some evidence that cones at or near the fovea centralis have and maintain much higher thresholds than do cones in the periphery. For example, the courses of dark adaptation using a retinal stimulus area of $\frac{1}{3}$-degree diameter situated 1 to 2 degrees from the small central red fixation source show one change and sometimes two very definite changes of threshold during the course of an hour's investigation. The total change of levels, the first of which may persist from ten to twenty minutes, is frequently as much as 0.75 log unit. Individuals vary in their responses; some subjects show little change of the threshold values of the cones which are reached about five minutes after exposure to a preadapting light source of 200 millilamberts for four minutes. A few presumably normal subjects show data indicating a cone threshold, under my method of testing, of 3.2 log units (micromillilamberts) for ten minutes after the assumption of this level, followed by a second and rapid change of level to 2.8 log units. This second level is maintained for fifteen to twenty minutes and may or may not show further change. Occasionally the third stage has been noted with a value of approximately 2.5 log units (micromillilamberts). The

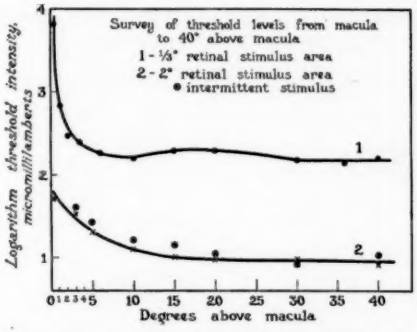


FIG. 22. Surveys of the threshold levels from the fovea to a region 40° above the macula using fields of $\frac{1}{3}$ degree and 2 degrees diameter, and subsequent to a period of dark adaptation of an hour's duration (Sheard).

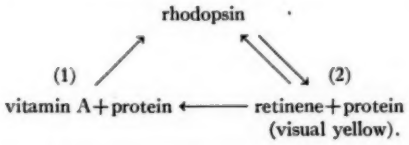
criticism may be raised that the fixation is not maintained constant and that, because of wandering or fatigue, more eccentric areas richer in rods are being stimulated. I have endeavored to guard against this possibility by suitable procedures and tests.

An interesting set of data in this connection is shown in Fig. 22. The two graphs are surveys of the threshold levels from the fovea to 40 degrees above the macula using a very small ($\frac{1}{3}$ degree) and larger (2 degrees) retinal stimulus area after the eye under test had been *dark adapted for nearly an hour*. Several sets of readings were made with the $\frac{1}{3}$-degree test area at 1, 2, 3, 4, and 5 degree positions, and then for every 5 or 10 degrees peripherally. The data of curve 1, Fig. 22, are of importance in that they show the maintenance of a high threshold on the part of the cones which are located in the fovea, with a rapid increase of threshold sensitivity of the receptor end-organs which respond in the macular area *in toto* and in surrounding retinal areas. As would be expected, curve 2 of Fig. 22,

in which a relatively large test field was used, shows the cone threshold to be of the order of 1 log unit above the rod threshold. It seems to me that there is no conflict between these findings and the data of many other investigators in the field of visual adaptation, for the reason that the angular diameter of test fields, intensities of preadapting light, and tests only on cones situated 7 to 10 degrees eccentrically to the macula have been employed by them.

8. Speed of Dark Adaptation and the Chemistry of the Rod Pigments

Sixty-five years ago Kühne concluded from direct observation of frog and rabbit retinas that visual purple may be synthesized by two procedures: a slow "neogenesis" from colorless initial stages following intense and prolonged irradiation, and a threefold or fourfold faster "anagenesis" from yellow initial stages following incomplete bleaching. As Wald and Clark (1937) pointed out, neogenesis and anagenesis may be identified with the formation of rhodopsin from vitamin A and from retinene, respectively. I am inserting Wald's equation again in order to discuss the dependence of the slow and fast synthesis of rhodopsin on the intensity and exposure to light. The chemical equation is:



The chemical equation permits the following predictions concerning dark adaptation: 1. Exposure of the dark-adapted eye to a short, intense flash of light should convert a large quantity of rhodopsin to retinene but little retinene should have had time to form vitamin A. Following such an exposure, dark adaptation should depend chiefly on reaction (2) and should be relatively rapid. 2. Protracted illumination of the eye should bring the visual cycle to a steady state; in this situation dark adaptation should depend principally on reaction (1) and should be relatively slow. 3. Since retinene is removed to form vitamin A in addition to rhodopsin, dark adaptation as it proceeds should depend more and more, and ultimately entirely, on reaction (1). The latter portions of dark adaptation should show the comparatively simple character of the slow process alone. In a general way, experimentation verifies these predictions.

Figures 23 and 24, taken from the reports of Wald and Clark, show that during the first few minutes dark adaptation of the rods is

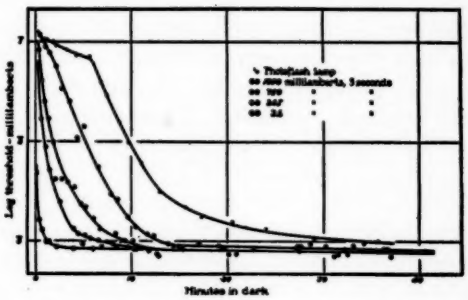


FIG. 23. Dark adaptation after short exposures to various intensities. The initial portions of rod dark adaptation increase in velocity as the light adapting intensity falls. (From Wald and Clark, 1937.)

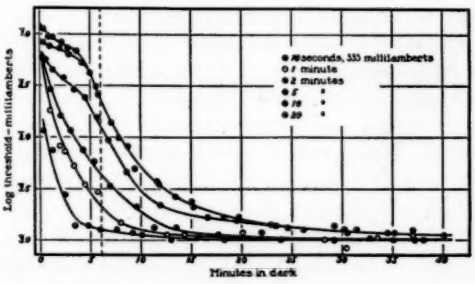


FIG. 24. Dark adaptation after various lengths of exposure to 333 millilamberts. As light adaptation proceeds, the visual threshold rises and independently the speed of rod dark adaptation decreases. (From Wald and Clark, 1937.)

rapid following short exposure to light but slower subsequent to long irradiation. As dark adaptation progresses, the slow process becomes more prominent than the rapid process and completely occupies the final stages of adaptation. Light adaptation displays similar duality. As the exposure to light of constant intensity increases, the visual threshold rises and, independently, the speed of dark adaptation decreases.

Haig (1941) investigated very carefully the course of rod dark adaptation as influenced by the intensity and duration of preadaptation to light. He concluded that an increase of the degree of light adaptation causes a decrease of the "slope of the subsequent rod dark adaptation function" and a displacement of the function to the right on the time axis. Over a wide range, these changes occur to the same extent irrespective of whether the increase of the degree of light adaptation is produced by raising the intensity or by prolonging the exposure. Within these limits the Bunsen-Roscoe reciprocity law applies to the intensity and duration of pre-exposure. Over a still wider range, dark adaptation follows the same course subsequent to a brief exposure to a bright light as it does after prolonged exposure to a dim light, provided the degree of light adaptation is the same in both instances, as indicated by identical initial thresholds of dark adaptation.

9. Use of Red Filters for Producing Dark Adaptation

On theoretical grounds it is known that the use of red light, and hence the wearing of red goggles or red spectral filters before the eyes, are effective in dark adaptation of the rod mechanism, which is, of course, responsible for night vision (Figs. 1 and 6). Many have made use of red goggles as a time-saver in becoming dark-adapted. For several years roentgenologists and x-ray technicians have used such goggles as a means of preparing their eyes for roentgenoscopic work. The goggles are worn for several minutes while engaging in ordinary office work before going into the dark room and during an intermission when the operator desires to leave the dark room. An excellent contribution to this phase of roentgenologic technique and the general topic of roentgenoscopes and roentgenoscopy is by Chamberlain (1942). In recent years some investigators have used such red goggles in connection with tests on visual adaptation of aviators and in other aeromedical problems.

Miles presented a paper on this subject in 1943 and concluded, in essentials, that, after adapting the eye to light in a standard manner and wearing the red goggles in a lighted room (4 millilamberts or practically 5 foot candles of illumination) for twenty-five minutes, the brightness thresholds were as low as or lower than when the subjects had been in complete darkness for the entire period. Dark adaptation levels made by using red goggles intermittently showed threshold curves that were as low as or lower than those made while the subject was in complete darkness. In Miles's opinion his data indicate that deep red light has a property of augmenting the rate of dark adaptation of the eyes beyond that provided by complete darkness during the first twenty-five to thirty minutes of adaptation.

My own experience is only partially in support of the conclusions of Miles. In general it has not been found that the levels were quite as good and never, as yet, has it been found that they were better with the use of standard and adequate red goggles than with complete darkness. However, it is certainly true that the technique of the use of red goggles provides a means whereby night or dark room duties can be interrupted at will by returning to lighted areas without the loss of dark adaptation.

However, I have obtained just as rapid dark adaptation and secured as great a degree of night vision through the use of neutral filters which transmitted relatively low amounts of incident light, as for example a transmittance of 10 percent. Various data indicate that the very considerable reduction of time required for dark adaptation which may be secured through the use of red goggles transmitting energy of wavelength greater than 600 millimicrons is caused by the fact that such goggles have lowered the brightness of the visual field, and is not due specifically to the character of the spectral region of radiant energy which is transmitted by them. In such tests it is obviously necessary that the various filters used should be adjusted to the same visual transmittance. In general, therefore, it has been my experience that the threshold levels after a given period of use of red goggles have not been as good as (although they have been very close to) the values obtained when the subjects were in total darkness and, furthermore, the use of neutral filters was as satisfactory as that of red goggles, indicating that it was the

reduction of brightness of field rather than of the character of the spectral transmission which was of prime importance.*

10. Effects of Ingestion of High Carbohydrate Meals and of Increased Metabolic Rates on the Courses and Threshold Values of Dark Adaptation

During the course of some investigations concerning the various factors which may influence or affect either, or both, cone and rod dark adaptations and the relative weight or importance of each factor, my colleagues and I found that the amount and type of food the subject had eaten, or perhaps the failure to eat, prior to the tests had an effect. The tests on fasting subjects seemed to indicate that the threshold levels were elevated. We made more than 100 tests on fifteen male subjects between the ages of seventeen and nineteen years prior to their induction into the military forces. Dark adaptation tests were made on all subjects at the same hour each morning subsequent to a fast from 7 P.M. the night before. Hence all tests made in the morning were started at 9 to 10 A.M. Upon completion of the visual adaptation tests, all subjects were given at noontime a meal containing 240 g of carbohydrate, 10.5 g of protein, and 9.5 g of fat. The food consisted of sweetened fruit juice, sliced grapefruit with sugar, oatmeal with sugar, one glass of milk and six pieces of stick candy. This type of meal can be duplicated easily and, furthermore, would be practical for use as a quick source of energy and of raising the level of blood sugar.

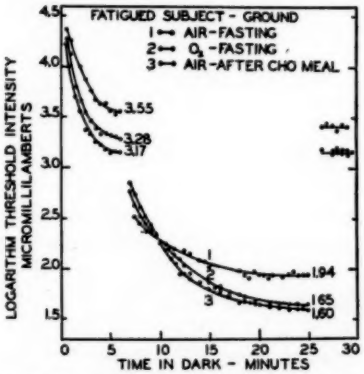


FIG. 25. Curves showing the effects of the breathing of oxygen and of the ingestion of a carbohydrate meal on a subject who evidenced fatigue. (From Sheard, Brown, and Wilson.)

For fasting subjects (for at least fifteen to sixteen hours) we found that the breathing of oxygen at ground levels not only increased the speed with which dark adaptation occurred but showed definitely lower final (twenty to thirty minutes) thresholds than were obtained when the subject was breathing air. Furthermore, for the fasting subjects, the courses and thresholds of dark adaptation were nearly the same after the ingestion of food irrespective of whether the subject breathed air or oxygen. Also, in a few instances in which there was evidence of fatigue we found that the thresholds of both cone and rod adaptation were raised (as much as 0.5 log unit or a decrease of sensitivity to a third) for a fasting subject and that, under these circumstances, the breathing of oxygen greatly improved the ability to see in the dark. The ingestion of a high carbohydrate meal by a subject restored the dark adaptation to its lowest (increased sensitivity) levels. There is much evidence that the ingestion of a high carbohydrate meal after fasting produces about the same improvement of the ability to see in the dark as does the breathing of oxygen by a fasting subject. In some cases the improvement produced by a high carbohydrate meal is even a little greater than that produced by the breathing of oxygen. Typical results (a full account of which will be published later) are given in Fig. 25. We have concluded that the amount and character of food ingested prior to tests on dark adaptation influence the course but more particularly the final level attained. A light meal of very high carbohydrate content appears to be quite effective in obtaining the best possible final cone and rod thresholds.

* Since the presentation of some portions of this paper before the Optical Society of America in March, 1943 and since the preparation of the first draft, a short article by Lowry has appeared in the literature concerning the effect of hue on dark adaptation. The results for six observers are given in the appended table and show the time required for each to reach a threshold of 0.035 microlambert after a one-minute exposure to a sensitizing field of 3.15 footlamberts brightness.

Observer	Red filter (seconds)	Neutral filter (seconds)
1	43	40
2	40	45
3	65	63
4	83	83
5	73	77
6	58	61

TABLE II. Values for dark adaptation and for serum vitamin A and carotenoids before and after administration of thyroid extract and of alpha-dinitrophenol. (From Patek and Haig, 1941.)

Subject	Test no.	Drug (T or D)	Daily dose, grains	Dark adaptation thresholds (log micromicrolamberts) Cones 2 min.	Rods 15 min.	Rods 40 min.	Cone/rod transition point, minutes	Serum vitamin A, I.U./100 ml	Serum carot. micrograms/100 ml	Basal metabolic rate
E.R., 55 years, female.	1	0		7.7	6.9	4.7	15.0	105	56	− 2
Rheumatoid arthritis.		13 T	2	6.3	4.0	2.7	4.5	93	42	+12
Renal stone removed 7 years ago.										
J.C., 64 years, male.	2	0		7.4	7.0	4.9	14.5	122	116	− 9
Cirrhosis of liver.		42 T	1	7.0	5.1	4.2	8.1	64	116	+12
	3	0		7.6	7.0	4.5	14.8	190	149	−11
		9 D	1.1	6.8	5.4	3.7	9.0	134	166	+ 9
	4	0		7.2	6.7	4.4	13.5	—	—	−11
		8 D	1.1	6.9	3.9	2.1	4.2	—	—	−10
J. S., 35 years, male.	5	0		8.1	6.7	5.0	12.8	0	458	−34
Cirrhosis of liver.		121 T	1–3	6.5	4.4	4.2	4.5	9	99	− 4
Myxedema.	6	0		7.6	5.6	4.8	9.8	70	200	−24
		56 T	1–3	6.8	4.7	4.2	5.5	56	158	−10

T, days of administration of thyroid.
D, days of administration of alpha-dinitrophenol.

There are various pieces of evidence from other sources which afford some support of, or substantiation to, the foregoing conclusions. In 1941, Patek and Haig reported their findings on the effects of the oral administration of thyroid or alpha-dinitrophenol on dark adaptation. These substances produced sharp changes of both rod and cone functions. Table II records some interesting and valuable data. Only the initial and final values for each test period are tabulated. The data show a considerable increase of speed of adaptation and lowering of visual thresholds after feeding of either thyroid or alpha-dinitrophenol. The type of response is similar to that observed for some persons who were given vitamin A but the response is accelerated. For example, patient J. C. required 250,000 to 500,000 international units of vitamin A daily for two months before normal values for dark adaptation were obtained. In some cases Patek and Haig stated that such values were reached in three to ten days after the feeding of either of the compounds mentioned previously. It is possible that the conversion of carotene may be facilitated by administration of thyroid. Such a possibility is suggested by the work of Fasold and Heidemann, and of Clausen and McCoord.

Why such substances as thyroid and alpha-dinitrophenol accelerate the rate of dark adaptation and lower the visual thresholds is none too clear. The fact that the two substances bring about the same response suggests that the visual changes, like the metabolic rate, are functions of general metabolism. Also the rate of regeneration of visual purple in vitro is increased by raising the temperature of the system (Zewi). Patek and Haig stated that there is no general correlation between the level of the basal metabolic rate and the speed or extent of dark adaptation. Their statement that, in a case of classical myxedema (B.M.R. −26 percent) and for normal subjects who had basal metabolic rates of −17, −19, −16, and −15 percent, respectively, the dark adaptation curves were normal in no wise disproves the statement, as I see the matter, that basal metabolic rates, especially in the extremes of high and low values, have an appreciable effect on dark adaptation. As might be expected, the improvement of speed and extent of rod adaptation subsequent to the administration of thyroid or alpha-dinitrophenol was greater than were the effects in the case of cone adaptation. In this respect the responses resemble those produced by vitamin A therapy and are in contrast to the equal (in general) lowering of the rod and cone thresholds observed to accompany recovery from anoxemia (McFarland and Evans, 1939; McDonald and Adler, 1939). These changes appear, therefore, to indicate that the utilization of vitamin A by the visual mechanism is facilitated by thyroid or alpha-dinitrophenol and that there is an alteration of the speed and extent

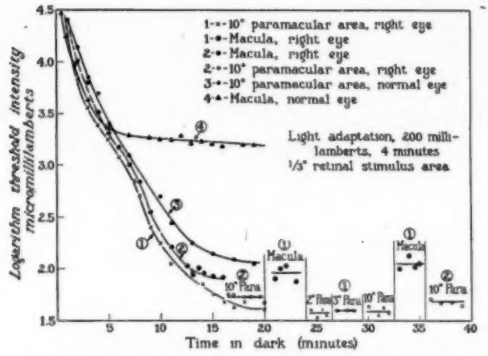


FIG. 26. Curves 1 and 2 are the course of dark adaptation for the rods and the cones, respectively, in a condition of achromatopsia. Curves 3 and 4 give the corresponding data in the case of an eye of normal acuity and adaptation. (From Sheard and Bair.)

of regeneration of visual purple rather than a local effect on the nervous mechanism.

Yudkin also investigated the effects of single doses of 100,000 international units of vitamin A alcohol, in gelatin capsules, given on an empty stomach. The effect on the dark adaptation curves was followed during the succeeding twenty-four to thirty-six-hour period. The subjects ingested food three hours after taking the vitamin A capsules. The results obtained varied somewhat; most commonly the improvement began in two hours and was maximal in ten hours, the curve of dark adaptation returning to its previous state in twenty-four hours.

II. CLINICAL CONDITIONS AND CONSIDERATIONS

From the statements and data which are contained in the preceding paragraphs regarding the biophysical and physiological measurements and responses of the rods and cones in visual adaptation, it is evident that there are three fundamental elements which may exert an influence on dark adaptation. These factors are:

(1) Pigment or pigments and the conditions that may affect the photochemical reactions and changes;

(2) Metabolism and nutritional state, both of the body and of the retina as a localized site of pigment and photochemical reaction;

(3) Neural and cerebral responses, which are exhibited so strikingly in anoxia and certain aeromedical problems. Without doubt, these neural and cerebral factors enter into many clinical syndromes to the point that, on occasion, I have remarked that I was unable to state with assurance whether the departures from the normal range of dark adaptation were due to pigmentary, metabolic, or neural changes or responses, or perhaps to a combination of all of them.

1. Rod and Cone Thresholds in Congenital Achromatopsia and Bilateral Macular Degeneration

Before passing into a consideration and discussion of vitamin A and dark adaptation and of various substances and drugs which may affect night vision, I am presenting some experimental data by Bair and me (as yet unpublished except in abstract form) on the cone and rod thresholds in conditions of congenital achromatopsia and of bilateral macular degeneration. We have had the privilege of making quite complete studies on three subjects. They showed $V = 6/60$ or thereabouts, total color blindness and central scotomata which were relative and not absolute. Perimetric fields were normal except for the central visual areas.

Total color blindness was discovered in 1880 and is a rare occurrence. It is a common saying that "all cats look gray in the dark," and to a person without any color sense (achromatopsia) all objects look gray by day as well as by night. The only difference between two color fields to the totally color-blind eye is difference of brightness. Even with daylight illumination, the visible spectrum as seen by the totally color-blind eye is just a grayish band of light with a brightness distribution very nearly corresponding to that of the twilight spectrum of the dark-adapted eye. The brightest region apparently exists in the interval between the wave-lengths 540 and 530 millimicrons, which is green for normal color vision. The parallelism between the vision of the light-adapted totally color-blind eye and the twilight vision of the dark-adapted normal eye, as shown for example by the curves in Fig. 26, is so close that there can be little doubt that the visual mechanism must be the same in the two cases. Indeed it is very probable that the real difference between a normal eye and one that is congenitally totally color-blind is that the latter lacks the so-called daylight (photopic) mecha-

nism altogether. In other words, the achromatic vision, either by day or by night, is rod vision pure and simple. The cones are, therefore, either lacking, inactive, or else functioning as rods only.

A person who is totally color-blind in general cannot see at all under a bright illumination such as is acceptable to most persons. He is photophobic, not so much because bright light is painful to him as because it is blinding. In total color blindness the visual acuity is poor, ranging from a fourth to a tenth of the normal visual acuity (V = 20/20) under the same external conditions. Furthermore, if congenital total color blindness is exactly similar to twilight vision, one would expect to find a central scotoma. In the three cases that we have investigated we have found central scotomata of relative rather than absolute character. If the central scotoma is absolute (or nearly so), it would be expected that such a person under test would have grave difficulty, if not the impossibility, of maintaining the direction of fixation. In fact, nystagmus nearly always accompanies total color blindness and there is little or no tendency to a coordination of the eyes which is so predominantly present in ordinary binocular single vision.

TABLE III. Dark adaptation in total color blindness and in macular degeneration. Values of the logarithms of the threshold levels (micromillilamberts) after various periods of dark adaptation. Retinal stimulus area (R.S.A.) = $\frac{1}{3}$ degree; light adaptation = 4 minutes to 200 millilamberts. (From Sheard and Bair, 1942.)

Subject	Area stimulated	Time of dark adaptation, minutes	Threshold level	Representative normal data*
		1	4.3	4.4
		5	3.35	3.4
	10 degrees	10	2.47	2.58
	above	15	2.00	2.16
	macula	20	1.78	2.08
		25		1.96
		30	1.60	1.94
A				
		1	4.15	4.20
		5	3.42	3.50
		10	2.56	3.34
	Macula	15	2.10	3.25
		20	2.04	3.20
		25	1.98	3.17
		30		3.09
		35	1.85	3.06
		1	4.35	4.4
		5	3.25	3.4
	10 degrees	10	2.25	2.58
	above	15	1.75	2.16
	macula	20	1.60	2.08
		25		1.96
		30	1.59	1.94
B				
		1	4.4	4.20
		5	3.4	3.50
		10	2.56	3.34
	Macula	15	2.10	3.25
		20	2.04	3.20
		25		3.17
		30		3.09
		35	1.98	3.06

* Average of 50 determinations on 10 subjects.

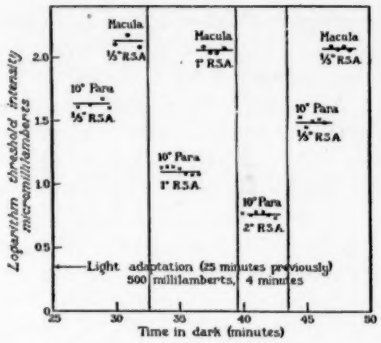


FIG. 27. Curves showing the levels of cone and rod adaptation with various sizes of field (R.S.A.) in a case of achromatopsia. (From Sheard and Bair.)

The curves of Fig. 26 are practically self-explanatory. Curves 1 and 2 are for the cone (macular) and rod (peripheral) dark adaptations in a condition of achromatopsia. Curves 3 and 4 show the corresponding data in the case of an eye that has normal acuity and adaptation. It is shown that the dark adaptation of the macular area is comparable, if not slightly superior, to the rod dark adaptation of the normal eye. Also in total color blindness as well as in macular degeneration, the rod dark adaptation is superior to that of the normal eye. In other words, the persons whom Bair and I tested were able to find the black cat in the black night looking straight ahead with greater certainty than either of the examiners using their best peripheral (rod) vision. Figure 27 shows the level of foveal (macular) adaptation and of a region 10 degrees paramacularly, the initial data being taken twenty-five minutes after recovery from exposure to a light source of 500 millilamberts intensity for four minutes. The findings in the macular region remain at the threshold of 2 log units (micromillilamberts) using a test area subtending an angle of $\frac{1}{3}$ or 1 degree in diameter.

If the retinal stimulus area was 2 degrees in diameter and was situated 2 degrees from the fovea (hence with a center 3 degrees from the fovea), the findings were of the same order as those given for the 10-degree paramacular area.

Table III gives the data in a condition of macular degeneration and shows that the dark adaptation in the macula is comparable (1.85 log units) to the rod dark adaptation (1.94 log units) of the average normal eye. At the end of thirty-five minutes of recovery in the dark, the threshold level of the macula is 1.85 log units (micromillilamberts) as compared with the average normal foveal cone adaptation value of 3.06 log units, a difference of 1.2 log units.

As I have stated, there arise the questions whether foveal fixation was maintained and whether it is possible that a secondary fovea may have developed and that it is the responses in a paramacular (and hence densely rod-populated) area which are recorded in our data. Perimetric and other tests and checks indicated that foveal fixation of the red light could be obtained and maintained.

Much speculation and theorizing have been indulged in regarding the shift of the wavelength distribution of brightness in total color blindness to the side of the spectrum which comprises the shorter waves. It is probable that the rod mechanism of the human eye is much earlier in origin than the cone mechanism, which is endowed with the faculty of color discrimination. Doubtless primitive vision was in black and white only. Phylogenetically, the cones in the periphery would be the oldest, those in the fovea the youngest in order of development. Schrödinger suggested that, while the old mechanism of vision was retained intact, a new mechanism was developed which acquired notably different properties. "According to this view, the occasional occurrence of total color blindness among mankind is to be considered as being merely a persistence or continuance in an earlier stage of development; in other words, as atavism."

2. Vitamin A and Dark Adaptation

The relations between vitamin A and dark adaptation depend on three known facts. In the first place, as has been shown by Wald (1935) and again by Hecht (1940), vitamin A is a constituent part of the photosensitive pigment (or pigments perhaps) of the rods and probably also of the cones. Secondly, it is an established clinical fact that many persons whose dark adaptation thresholds are abnormal are improved in their ability to see in the dark through the administration of vitamin A. Among the earliest contributions on the subject of functional night blindness due to vitamin deficiency in man are those of Aykroyd (1930) and Tansley (1931). The most frequently cited publications are the contributions of Edmund and Clemmesen (1936); Jeans and Zentmire (1936); Wald, Jeghers, and Arminio (1938); Hecht and Mandelbaum (1939); and Lewis and Haig (1940). Thirdly, night blindness can be produced in some instances by feeding the subject a diet deficient in vitamin A (Jeghers, 1937; Booher, Callison, and Hewston, 1939; Steininger, Roberts, and Brenner, 1939).

All relevant investigations are based on one or more of these facts. Many criticisms that have been directed against the fundamental truth of these three statements are irrelevant because of inaccurate methods of testing dark adaptation (see Dow and Steven, 1941, for a detailed criticism of methods) or the subjects and conditions of the tests are controlled inadequately, with a total disregard, in some instances, of the role that various physical and physiological factors may play. If no organic disease of the eye, such as retinitis pigmentosa, is present, measurements of dark adaptation, when made under critically standardized and controlled conditions, can be used as aids in the determination of avitaminosis A, which may be produced by a lack of the vitamin in the diet, by a functional disarrangement of the transfer of the vitamin from the diet to the retina, by a lack of reserves in the liver, or by unavailability of reserves.

In 1938 Hecht and Mandelbaum investigated the relation between vitamin A deficiency in the food and the thresholds of rod and of cone dark adaptation. They adopted the standard apparatus and procedure of Hecht and Shlaer, involving preadaptation to 1500 millilamberts for three minutes and measurements with extreme violet light of the adaptation of a retinal area 3 degrees in diameter situated 7 degrees nasally

from the fovea. They made measurements of the dark adaptations of four normal young men, first of all on their regular and adequate diet and then on a diet containing only about 150 units of vitamin A each day in order to find out whether, under these circumstances, the behaviors of the rods and cones followed parallel courses.

Figure 28 contains several curves concerning the dark adaptation made with one subject during different stages of the deficient diet. In all curves it is evident that there are two portions: the first section, occurring fairly rapidly, represents cone adaptation; the second section, which commences after about eight minutes in the dark, represents rod adaptation. It is evident that both the cone and the rod function are affected by dietary deficiency of vitamin A. The effects, however, are greater on the rod function than on the cone function. The cone-rod transition point remained the same (seven to eight minutes) during the thirty-five days of the investigation. This was found to be true for all four subjects tested by Hecht and Mandelbaum. On the other hand, under the same physical and environmental conditions, there is a displacement of the cone-rod transition point to as long as fifteen to twenty minutes for patients who have cirrhosis of the liver, with a gradual shift toward the normal interval of time if the treatment with vitamin A is effective.

Figure 28 shows that as the supplies of vitamin A or other substances (from which the rhodopsin of the rods and pigments of the cones are produced in, or are present in, the retina) are reduced there is a general rise of the level of the thresholds of both rods and cones. The parallel behavior of rods and cones is shown conclusively in Fig. 29. These records show the results (from Hecht and Mandelbaum) on two subjects who were given supplements of vitamin A subsequent to the cessation of a diet very low in carotene and vitamin A and after the return to a normal diet plus 50,000 units of vitamin A daily. The data of J. M. (Fig. 29) are about average for the four subjects; L. W. represents the extreme effects. Each point in Fig. 29 is the final threshold either of the cone section or of the rod section of such curves as those shown in Fig. 28. The thresholds of the last two tests in which the subject was on a normal diet are shown above the diagonally shaded area on the time axis; the clear area records the duration of the dietary deficiency, while the blackened area represents the subsequent normal diet fortified with vitamin A.

The curves of Fig. 29 show that, as measured by dark adaptation, the presence of dietary avitaminosis became evident after the first day

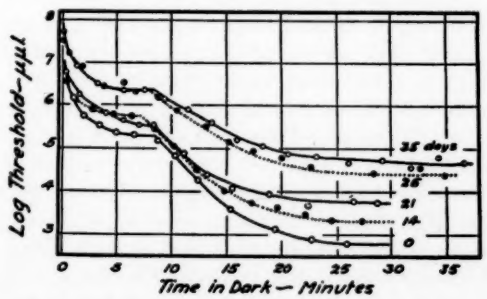


FIG. 28. Dark adaptation curves made at various periods during a diet deficient in vitamin A. (From Hecht and Mandelbaum, 1938.)

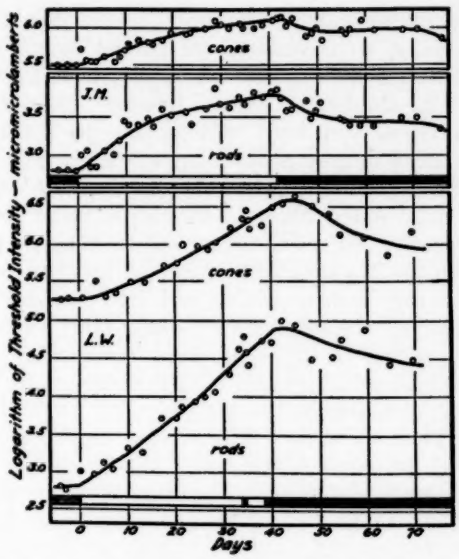


FIG. 29. Final rod and cone thresholds of two subjects on a vitamin A controlled diet. The diagonally shaded part represents a normal diet; the clear part represents a vitamin-free (practically) diet, while the blackened part indicates a normal diet with 50,000 units of vitamin A daily. (From Hecht and Mandelbaum, 1938.)

of the absence of vitamin A in the diet. Obviously, on account of the normal day-to-day variation, no reliance can be placed on such a single early measurement. However, the rise of threshold continued steadily and in about a week of vitamin A deficiency in the diet the rod and cone thresholds had risen beyond their daily normal fluctuations. At the end of about fourteen days the rod threshold had risen beyond the commonly encountered range of levels of presumably normal persons and after twenty-one days or thereabouts the levels, both for the cones and for the rods, were higher than the extreme which is found to exist in only 3 percent of a normal population.

The return to a normal diet supplemented with 50,000 units of vitamin A daily resulted in a rather slow recovery. In contrast to these results of Hecht and his colleagues concerning the effects of high vitamin A diets on dark adaptation subsequent to a dietary deficiency are the spectacular recoveries of the visual function reported by Edmund and Clemmesen (1936) and by Jeghers (1937) and the almost immediate return to normal reported by Wald, Jeghers, and

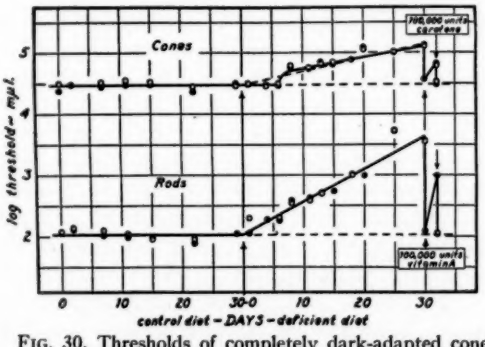


FIG. 30. Thresholds of completely dark-adapted cones and rods (cone and rod "plateaus") during thirty days of optimal vitamin A nutrition (left portion) and during thirty days on a diet deficient in vitamin A (right portion). Open and closed circles show the thresholds of the right and left eyes, respectively. On the thirtieth day of deficient diet, vitamin A was administered: the course of the curve is shown in Fig. 31, *A*. On the thirty-second day the subject was again hemeralopic and was given carotene. (From Wald, Jeghers, and Arminio, 1938.)

Arminio (1938). Differences exist in the experimental procedures and dietary regimens of all these workers; in some cases there is a decrease of, and in other instances a supplementary addition of, vitamin B or other vitamins. Such modifications and departures, insignificant as they may appear at first glance, are probably of importance and exercise a considerable effect either as compensatory agents or in some undetermined manner.

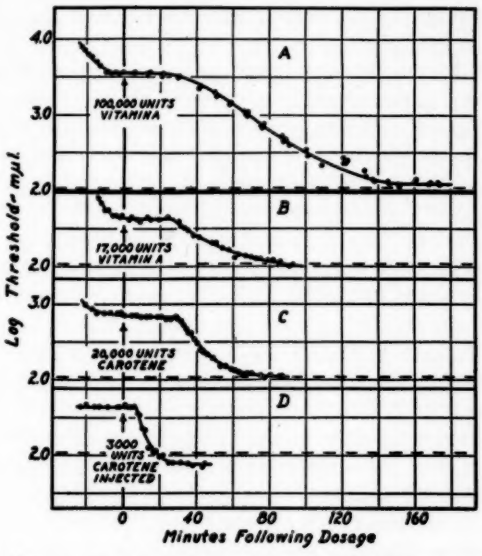


FIG. 31. Relief of hemeralopia by administration of vitamin A and carotene. Measurements preceding administration show terminal portions of dark adaptation. In each section of the illustration, a broken line indicates the average threshold of the normal dark-adapted eye. (From Wald, Jeghers, and Arminio, 1938.)

Figure 30, taken from Wald, Jeghers, and Arminio (1938), shows the thresholds of completely dark-adapted cones and rods (that is, cone and rod plateaus, so-called) during thirty days of optimal administration of vitamin A (left portion) and during thirty days on a diet deficient in vitamin A (right portion). The open and closed circles show the thresholds of the right and left eyes, respectively. Vitamin A was administered on the thirtieth day of the deficient diet. The course of the response to treatment is shown in Fig. 31, *A*. On the thirty-second day the subject was again hemeralopic and was given carotene orally. After a latency of only thirty-eight minutes the rod threshold returned from a level of 3 log units (micromillilamberts) to a normal rod range slightly above 2 log units. In Fig. 31, measurements preceding administration of vitamin A show the terminal portions of

the dark adaptation. The threshold is definitely hemeralopic at the beginning of each test; after the oral administration of vitamin A or of carotene (*A* to *C*, Fig. 31) or after their intramuscular injection (*D*) the thresholds return to a final level well within the normal day-to-day range. The course of dark adaptation of the rods descends to normal about twice as rapidly after the administration of carotene as after administration of vitamin A. Wald conjectured that the difference of effectiveness might be due to the greater efficiency of the absorption and transport of carotene. Wald and his associates all confirmed the assertion that the behavior of the cones parallels that of the rods; hence vitamin A appears to be the precursor of the visual pigments of the cones as well as of the rhodopsin of the rods.

A major portion, to say the least, of the researches which were published concerning vitamin A deficiency and dark adaptation from 1930 to 1940 laid stress on (1) the similarity of effects on cone and rod functions in vitamin A deficiency, (2) the relative ease with which dietary vitamin A deficiency could be produced, and (3) the inference, if not the direct statement, that there was a fairly high incidence of vitamin A deficiency (for example, Jeans and Zentmire, 1934–36). There was not agreement about the period involved or the speed of recovery subsequent to the administration of vitamin A or carotene; some investigators found little improvement in months, other workers found the dramatic thirty-eight minute recoveries. As we now know and as we might have suspected, both possibilities exist, with the bulk of clinical conditions showing very considerable improvement after a few weeks of adequate treatment.

In 1939 Steffens, Bair, and I published an account of our investigations on visual adaptation of normal adults placed on diets deficient in vitamin A. In brief, my colleagues and I found that, in three normal adult subjects (Fellows on the Mayo Foundation) the courses and the thresholds of dark adaptation of the macula (cones) and of regions 10 degrees above the macula (rods) were not affected significantly (less than 0.4 log unit) during the course of a dietary regimen very low in vitamin A content (100 to 300 international units daily) for periods of 42, 160, and 190 days, respectively. Figure 32 contains composite curves of the values of the dark adaptation, using a $\frac{1}{3}$-degree stimulus area and a previous light adaptation of 160 millilamberts for three minutes, for the macula (curves 1, 2, and 3, after ten minutes of dark adaptation) and for an area 10 degrees paramacular (curves 4, 5, and 6, after twenty minutes of dark adaptation) during the course of the investigations of three subjects on a diet deficient in vitamin A, and after the resumption of normal diets supplemented with 80,000 units of vitamin A for seventy days in the case of one subject. (Curves 1 and 4, Fig. 32, are for subject 1, curves 2 and 5 for subject 2, and curves 3 and 6 for subject 3.) The greatest elevation of level of dark adaptation was 0.6 log unit at the end of 120 days on the vitamin A deficient diet (subject 2).

In general, the thresholds, instead of showing an increase throughout the course of the period in which the diet was deficient in vitamin A, tended to decrease in the later part of the period and to assume the same levels as had been observed when the subjects were initially on the normal, adequate diet. Also, after the cessation of the deficient diet and during the period of normal diets supplemented with large doses of vitamin A, the threshold values were not affected. Various experimental checks and procedures were

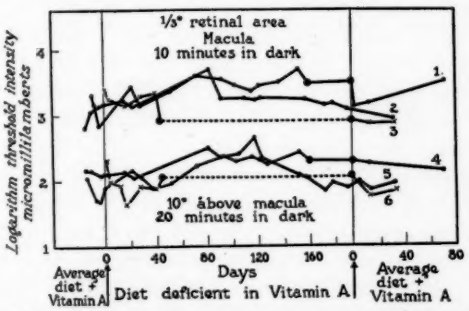


FIG. 32. Composite curves of the courses of dark adaptation for the macula (curves 1, 2, and 3) and for an area ($\frac{1}{3}$-degree diameter) 10 degrees above the macula (curves 4, 5, and 6) during the course of investigation of three subjects on a diet deficient in vitamin A and after the return to normal and fortified diets. Curves 1 and 4 (subject 1); curves 2 and 5 (subject 2), and curves 3 and 6 (subject 3). Macular (cone) thresholds are given after ten minutes of dark adaptation; paramacular (rod) thresholds after twenty minutes of dark adaptation. (From Steffens, Bair, and Sheard, 1940.)

used and it was concluded that, for certain normal adults, considerable periods (as great as 180 days) of deficiency of intake of vitamin may be maintained without producing any significant changes of rod or cone sensitivity. I might say that the data in Fig. 32 could be interpreted to indicate that there was a definite rise of thresholds of both the rods and the cones to the point of mild hemeralopia during the 80- to 120-day period and that the subsequent improvement of thresholds and ultimate return to the original normal levels were caused by some compensating mechanism.

As a matter of fact, one of the subjects who continued on the experiments for nearly nine months showed an extreme pallor of the skin during the last forty to sixty days of the 189 days of vitamin A deficient diet and there is a fair amount of evidence that the skin and fatty tissues were depleted slowly of their vitamin A stores. Biopsy of the skin of the person under discussion showed that, at the conclusion of the 189 days of vitamin A deficient diet, there were (1) relative and absolute hyperkeratosis and keratotic plugging of hair follicles, (2) acanthosis and intracellular edema present in moderate amount in the prickle-cell layer, (3) pyknosis of some of the nuclei and an increased amount of melanin in the basal-cell layer, and (4) absence of sebaceous glands. After this subject had been placed on the normal diet supplemented by 80,000 units of vitamin A daily for seventeen days, another biopsy was performed. The second biopsy showed that the skin was essentially normal except for occasional keratotic plugs in the hair follicles. These findings on the skin furnish evidence that the earliest signs of vitamin A deficiency may not always be shown by tests of the dark adaptation levels but by microscopic examinations of sections of the skin. This conclusion is in agreement with the clinical experience of certain other investigators (Frazier and Hu, 1936; Youmans and Corlette, 1938). Such cutaneous changes as I have outlined have been believed to occur generally in the late stages of A avitaminosis.

The most recent contribution on the effects of vitamin A depletion on young adults that I have seen is the excellent paper by Brenner and Roberts (1943). They placed five young adults, three women and two men, on a vitamin A low diet for seven and a half and four and a half months, respectively. At suitable intervals measurements were made of dark adaptation, vitamin A and carotene in the blood, total white cell counts and differential blood counts, and changes in the structure of the skin and of the conjunctiva. None of these measurements showed any definite changes from those found prior to the administration of the vitamin A deficient diet. It would appear, therefore, that such persons had sufficient stores in the liver and other tissues to withstand the effects of the depletion diet or that such signs as significant changes of dark adaptation are not merely the result of uncomplicated vitamin A deficiency. Furthermore, these data and conclusions of Brenner and Roberts support the conclusion of Steffens, Bair, and myself that there is little evidence to show that any considerable part of the population is suffering from a vitamin A deficiency such as has been reported in the literature from time to time.

Comment

Whether or not the findings and conclusions of Jeans and Zentmire are to be accepted, it is to be said that these investigators stimulated clinical interest in the possibility of employing dark adaptation as a method of detecting subclinical states of vitamin A deficiency. By means of a biophotometer, which is a modification of the Birch-Hirschfeld photometer, Jeans and Zentmire found a disturbance in 21 percent of a group of 213 children in a children's hospital in Iowa. About half of the children who showed relatively low rates of dark adaptation and, therefore, relatively high values of threshold intensities were placed on a high vitamin diet including cod liver oil and, after a period averaging twelve days (from four days to six weeks), all were reported to have demonstrated an improvement of dark adaptation. However, Maitra and Harris (1937) in England, Mutch and Griffith (1937), and Park (1935) in Oklahoma concluded that the recorded incidence of vitamin A deficiency is likely to be high if the methods of operation of photometers and the standards assumed by Jeans and Zentmire, and by others also, are employed. Also it is felt that,

although some relationship between the photometric readings and the state of vitamin A nutrition may exist, the relationship is not close enough to warrant the use of the test as a means of diagnosing subclinical vitamin A deficiency (Schuck and Miller, 1938; Youmans, Corlette, and Corlette, 1937; Palmer and Blumberg, 1937). Furthermore, it has been impossible in many cases to correlate the variations of photometric determinations of dark adaptation with the content of vitamin A in the diets, or again of the level of vitamin A in the blood.

Jeghers tested 162 medical students and found fifty of them who were subnormal in their dark adaptation according to the biophotometric determinations and the standards of normal visual adaptation which were used. Fifteen of these students had cutaneous symptoms. The differences between the subjects examined by Jeghers and those tested by other investigators may be real. However, it now appears likely that those differences are exaggerated by the apparatus and method used by such investigators as Jeghers, and Jeans and Zentmire. In 1939 my colleagues and I found that if the method of making determinations of thresholds is modified (by the occlusion of one eye, adequate fixation of the upper spot of the quincunx and the use of the central area of the quincunx as the stimulus area) the courses of dark adaptation and the thresholds obtained with the biophotometer are comparable to the data obtained with satisfactorily standardized ensembles and procedures. Isaacs, Jung, and Ivy (1938) have shown that the statistical error involved in the use of the biophotometer is high. Hecht and Mandelbaum stated: "The biophotometer does not incorporate in its structure and recommended procedure many of the critical specifications for measuring dark adaptation . . . and fails to link its results with physiologic knowledge." It seems to many investigators that too much reliance has been placed on the measurement obtained very soon (as quickly as twenty seconds) after the subject has been in the dark.

Other investigators have used the threshold of the dark-adapted eye in a manner and in a connection which make the procedure more specific, as, for example, the determination of vitamin A lability or of the vitamin A of the blood. Such methods have indicated thus far a very low incidence of vitamin A deficiency. Lindqvist (1938) found abnormally high visual thresholds (reduced light sensitivity) accompanied by abnormally low concentrations of vitamin A in the blood serum in less than 5 percent of a group of hospitalized patients who were suspected particularly of having vitamin A deficiency. Lewis and Haig (1940) tested a group of 144 children in New York City and concluded that only one child showed definite evidence of vitamin A deficiency as judged by data on dark adaptation. When one considers that many of the children, as Lewis and Haig pointed out, came from very poor homes and that, furthermore, many of them had been suffering from a febrile disease, the conclusion seems warranted that "vitamin A deficiency, on the basis of dark adaptation, is an uncommon disorder in children in New York City." To say the least, these results from a large city are in marked contrast to the results occasionally reported concerning the children of rural communities. And yet some of us have been amazed to discover from time to time that the country children and not the city dwellers suffer from various types of deficiency disease, possibly for the reason that the rural dwellers carry on without much of the governmental assistance afforded to city dwellers during times of depression and maintain their independence and social stability even to the point of considerable sacrifice.

Interestingly enough, Steven and Wald (1941) reached conclusions similar to those stated by Lindqvist (1938), Lewis and Haig (1940), Steininger, Roberts, and Brenner (1939), and others, after their measurements on 353 persons in Newfoundland and Labrador, areas from which epidemic night blindness had been reported. They employed a vitamin A labile threshold as a criterion of deficiency—a threshold such that, whatever its initial value, there would be an improvement of at least 0.3 log unit within two weeks of regular daily supplementation of diet with cod liver oil containing 15,000 international units of vitamin A. Vitamin A labile thresholds were found in 9.7 percent and clinical night blindness in at most 3 percent of the subjects. No xerophthalmia or keratomalacia was encountered. Also, beriberi is endemic in

northern Newfoundland. It was found that vitamin A labile thresholds and beriberi are mutually independent. For subjects who had both deficiencies, brewers' yeast did not lower the threshold, while vitamin A alone was found to be as effective as vitamin A and brewers' yeast combined.

In a report in 1941 from Strakosch of the Minneapolis General Hospital it was stated that eleven patients of the 115 persons examined gave subnormal photometric readings. Josephs, Baber, and Conn (1941) studied the vitamin A content of the blood and the dark adaptation in various socioeconomic groups and found that only in severe and prolonged deficiency of vitamin A were these measurements affected. The failure for six months or more of controlled diets, low in vitamin A, to result in any of the specific signs of deficiency is also indicative that the period of depletion was not ample. Theoretical calculations on the average content of vitamin A in the human liver (for example, Abels, Gorham, Pack, and Rhoads, 1941) and the assumption that the rate of use is approximately 2000 U. S. P. units per day indicate that it would take from one to two years for the liver to lose its entire store of vitamin A even if there were no vitamin A present in the diet. And again, Lewis and Haig (1939) investigated the requirements of vitamin A as determined by dark adaptation and concluded that infants receiving only 135 to 200 units of vitamin A had as good a threshold for vision in the dark as infants receiving an average diet supplemented by 17,000 units of vitamin A.

From the foregoing rehearsal of experimental findings and conclusions, as well as from a wealth of investigations which it is impossible to quote, it is evident that there is much that needs to be done on the metabolism of vitamin A to elucidate the conflicting, if not at times confusing, evidence.

3. Dark Adaptation in Cases of Avitaminosis A, Hyperkeratosis, and Cirrhosis of the Liver

While it is true that dietary avitaminosis A may be produced readily in some few persons but with considerable difficulty, if at all, in the majority of the population of the United States and while it is true that there is much evidence indicating the presence of other factors when dietary vitamin A deficiency is produced as shown by tests on dark adaptation, it is also true that the response to treatment with large doses of vitamin A concentrates and foods in frank conditions of deficiency of vitamin A is slow and recovery may involve weeks or months. Again, the intake of vitamin A in a normal diet may not be utilized by gastro-intestinal absorption and the vitamin A ingested may be excreted from the body. As a matter of fact, it may well be that, in any normal person, 99.95 percent of the vitamins may be passed on and into the sewer. Or again, in conditions of malnutrition due to diseases such as ulcerative colitis, it is entirely possible that there may not be enough retention, absorption, and utilization of vitamin A and other essentials of the ingested food to maintain adequate well-being with normal retinal functions and normal cerebral recognition and responses.

Furthermore, the tissues of the skin need carotene and vitamin A for their nourishment; indeed a factor in the production of the dominant yellow hue of all races is carotene in the skin. Hence it is easily understandable that cutaneous abnormalities or diseases may be associated with inadequate vitamin A in the blood and tissues and, since the retina is but a highly specialized skin, high levels (decreased sensitivity) of dark adaptation may occur simultaneously, or the one precede the other, or perhaps only one or the other be present. Hence it should be expected that dark adaptation levels would be high in cases of hyperkeratosis and pityriasis rubra pilaris.

Moreover, the liver is without question the great storehouse of vitamin A. Therefore, in diseases of the liver, such as cirrhosis, it would be expected that evidence would be found clinically indicative of a deficiency of vitamin A through departures from the normal ranges of rod and cone adaptation.

I. Dietary Avitaminosis A

The curves given in Fig. 33 show the changes of the levels of the thresholds, at the macula and

in an area 10 degrees above the macula, which were obtained from a patient who had been on a diet deficient in calories, proteins, and vitamins for about nine months to a year prior to examination at the Mayo Clinic. The patient gave a history of severe headaches associated with nausea and vomiting over a period of four years. Special diets had been prescribed and she had reduced her intake further by voluntary restriction. The course of dark adaptation in both the macular and the paramacular region (curves 1,

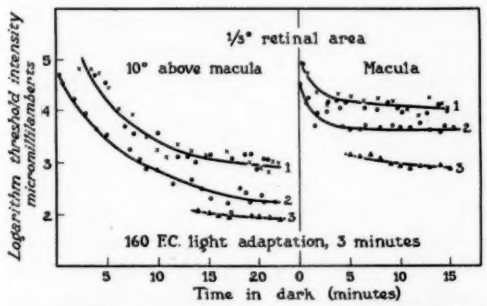


FIG. 33. Curves showing the course of dark adaptation in a subject who was poorly nourished and who had been placed on a restricted diet for many months. The areas investigated were the macula and 10 degrees above the macula, using a ⅓-degree stimulus area after a previous light adaptation to 160 footcandles for three minutes. Curves marked 1 (macular and paramacular regions) show the values of the threshold levels as initially obtained (indicated by ●), and as obtained eight days later (indicated by ×) after the subject had been placed on normal diets supplemented with high daily intake of vitamin A. Curves marked 2 (for both macular and paramacular areas) show the values of the light thresholds of the subject after being placed on an adequate diet with large daily intake of vitamins over a period of about four months. Curves marked 3 contain representative data of a normal subject. After four months on a normal diet, richly supplemented with vitamins, there is a reduction of the threshold levels (at the 20-minute point of the course of dark adaptation) of about 0.8 log unit (to a sixth) in the paramacular region chosen and approximately 0.5 log unit (to a third) at the macula. (From Steffens, Bair, and Sheard, 1940.)

Fig. 33) shows that the levels of the thresholds are about 1 log unit above the average normal level (curves 3, Fig. 33). Subsequent to the initial determinations the patient was placed on well-balanced diets supplemented by large daily doses of vitamin concentrates. During the first ten days on the regimen rich in vitamin content, there was no change of response in either the macular or the paramacular readings, as is indicated by the readings marked × on curves 1, Fig. 33. After four months, during which the adequate diet was supplemented by vitamin A, the measurements of the light thresholds gave the data shown in curves 2. There is a reduction of about 0.8 log unit in the paramacular area and approximately 0.5 log unit at the macula. The tendency toward restoration to normal thresholds appears to be greater, as a general rule, in paramacular (rod) areas than at the macula. In other clinical cases in which there were abnormally high levels, we have found that there may be a restoration to normal values in the light thresholds of the peripheral portion of the retina accompanied by only partial restoration of the thresholds of the macula. In the particular case under discussion (Fig. 33) normal levels of rod adaptation (that is, approximating curve 3) were found after the patient had been on a proper dietary regimen for a year. The cone level did not assume the value indicated as normal; at the end of a year's treatment the thresholds were of the order shown in curve 2, Fig. 33.

The case on which I have given data shows evidence of visual dysaptation and rather slow subsequent return to normal levels after high vitamin A intake. In other instances, however, the abnormally high levels may be maintained and little or no change be produced by adequate diets continued for several months. And again, somewhat in disagreement with both types of findings are those cases in which little change is found in the thresholds during the first week to possibly three weeks, followed by restoration of light thresholds to normal values in the course of the subsequent twenty-four to forty-eight hours, when an adequate diet was supplemented with 50,000 to 100,000 units of vitamin A daily throughout the course of the tests. Whether massive doses such as are recommended by some investigators would have proved more efficacious than the doses actually given is in the land of the unknown answers.

II. Follicular Hyperkeratosis and Pityriasis rubra pilaris

Table IV contains data on the values of the threshold levels in a representative case of follicular hyperkeratosis. Although the patient did not give a history of night blindness (and

TABLE IV. Dark adaptation in follicular hyperkeratosis. Values of the logarithm of the threshold levels (micromillilamberts) after various periods (minutes) of dark adaptation on days subsequent to treatment. Retinal stimulus area = ⅓ degree. Light adaptation = 3 minutes to 160 footcandles. (From Steffens, Bair, and Sheard, 1940.)

Time (minutes) dark adaptation	Days 1	3	10	14	18	23	Difference between initial and final values (log unit)	Representative normal data (average 50 different determinations on 3 subjects)
				I. Area 10 degrees above macula				
1	4.45	4.30	3.95	3.90	3.85	3.91	0.54	4.05
5	3.85	3.75	3.35	3.30	3.35	3.23	0.62	3.35
10	3.32	2.68	2.58	2.65	2.55	2.38	0.94	2.58
15	2.85	2.42	2.32	2.40	2.20	2.12	0.73	2.26
20	2.78	2.40	2.10	2.15	1.82	1.78	1.00	2.08
				II. Macula				
0.5	4.92	4.75	4.55	4.75	4.31	4.28	0.64	4.15
5	4.15	3.90	3.63	3.68	3.35	3.41	0.74	3.42
10	4.10	3.90	3.60	3.70	3.42	3.35	0.75	3.34
15	3.95	3.88	3.65	3.68	3.43	3.40	0.55	3.25
20	4.05	3.75	3.53	3.68	3.50	3.32	0.73	3.20

many such patients seem wholly unconscious of their impaired vision in the dark) it seemed desirable to determine the courses of dark adaptation because of the extensive keratinization of the follicular orifices. The levels obtained before treatment were found to be 0.4 to 0.85 log unit higher than the average values obtained from fifty different sets of determinations on three normal subjects. After the initial determinations of dark adaptation, the diet of the patient was fortified with food of high vitamin content, including fats. Carotene in oil was given orally. Inunctions of cod liver oil, about 150 cc daily, were administered to the entire body. About 150,000 units of vitamin A were administered daily. Other localized treatments with salicylated creams and the artificial induction of sweating by administration of pilocarpine were used. Determinations of the courses and levels of dark adaptation were made on various occasions during a period of twenty-four days. The values of the thresholds given in Table IV show a gradual return to normal levels while the patient was undergoing treatment. There is a decrease of from 0.5 to about 1 log unit in the values of the thresholds at various periods (minutes) of dark adaptation during the course of treatment.

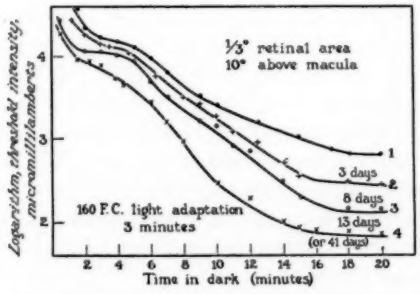


FIG. 34. The course of dark adaptation 10 degrees above the macula in a case in which the condition was diagnosed clinically and histologically as pityriasis rubra pilaris, on different days after treatment. Curve 1 was obtained on the day on which treatment was instituted. Curves 2, 3, and 4 represent the courses of dark adaptation which were obtained on the third, eighth, and thirteenth (also on the forty-first) days of continuous treatment. Curve 4 closely approximates the threshold values of a normal subject. (From Brunsting and Sheard, 1941.)

In the examination of three persons whose condition was diagnosed as pityriasis rubra pilaris, the preadapting light was 160 millilamberts exposed for three minutes. Under these conditions of light adaptation and test, the usual run of subjects who do not complain of photophobia or difficulty in seeing at night do not exhibit any initial stage of cone adaptation that can be determined definitely. In the cases which Brunsting and I studied, there were definite signs of abnormally high levels of dark adaptation with a period of cone adaptation which might continue for approximately five minutes before the onset of the course of rod adaptation. This phenomenon is exhibited in the initial

portions of the curves of Figs. 34 and 35. As the level of rod adaptation approaches the normal level there is a definite decrease of the period of cone adaptation (curves 1 and 4, Fig. 35). All the cases indicated the presence of vitamin A deficiency. After treatment, there was progressive lowering of the threshold levels, which was accomplished with comparative rapidity in some instances.

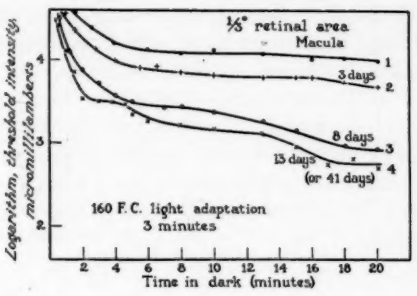


FIG. 35. The course of dark adaptation at the macula, with the same procedures carried out on the patient shown and discussed in connection with Fig. 34. (From Brunsting and Sheard, 1941.)

III. Cirrhosis of the Liver

In addition to inadequate intake of vitamin A, there are many conditions which may lead to a deficiency. Fever, general infection, rapid growth, elevated basal metabolic ratè, pregnancy, lack of bile or pancreatic secretions, changes of the gastro-intestinal mucosa, disturbances of the gastro-intestinal tract, or hepatic disease may prevent or hinder the proper absorption or storage of vitamin A. Hepatic disease is mentioned frequently as a cause of vitamin A deficiency. Clinical investigations have been somewhat inconclusive because of the occurrence of jaundice which, with the resultant faulty absorption of the vitamin due to lack of bile in the intestinal tract, may lead to vitamin A deficiency. Furthermore, the diets of many patients suffering from cirrhosis of the liver are restricted. Pathologic and experimental data concerning the relation between vitamin A deficiency and altered hepatic function have been inconclusive. Hence it is essential that tests be made to determine whether a deficiency of vitamin A occurs among patients suffering from hepatic disease whose intake of the vitamin is adequate and who are not jaundiced.

The reports of Patek and Haig (1939) are conclusive in that they show a definite relationship between the thresholds of dark adaptation and

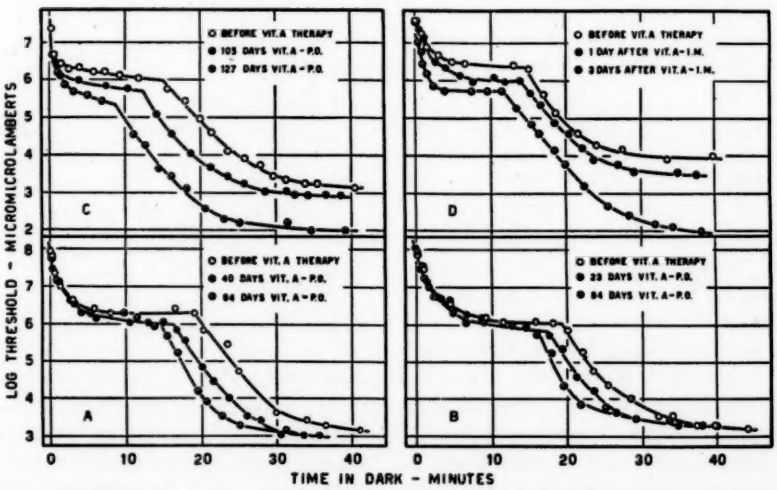


FIG. 36. Changes in the dark adaptation curves and thresholds of four subjects suffering from cirrhosis of the liver during the administration of vitamin A. The upper curves from cases *C* and *D* reveal high cone and rod final thresholds together with delay of rod adaptation. The lower curves of cases *A* and *B* show normal final thresholds but abnormal delay at the cone-rod transition point. In all cases presented, there is improvement of visual adaptation subsequent to vitamin A therapy. (From Patek and Haig, 1939.)

the administration of vitamin A to patients suffering from cirrhosis of the liver. Figure 36 shows the changes of dark adaptation of four patients during vitamin A therapy. Some of the data (cases *C* and *D*) reveal initially high cone and rod final thresholds together with delay of rod adaptation. Other experimental results (cases *A* and *B*) reveal normal final thresholds but abnormal delay at the point of cone-rod transition. (A white preadapting brightness of 4700

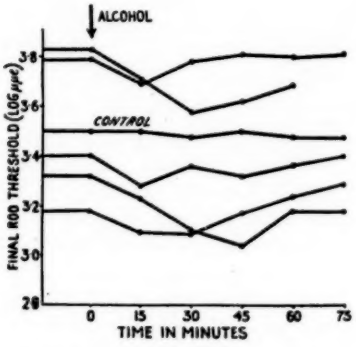


FIG. 37. Final rod thresholds of five subjects who were given 20 ml of alcohol and of a control subject without alcohol. (From Yudkin, 1941.)

millilamberts was viewed by the subject for four minutes; hence the normal period of cone adaptation would be ten to twelve minutes prior to the commencement of rod adaptation.) The results of Patek and Haig's investigations show that abnormal dark adaptation occurred in about four-fifths of the cases of cirrhosis of the liver. In certain cases both elevation of the final rod and cone thresholds and delay of rod adaptation occurred; in most instances the latter change only was present. These changes were not related to jaundice. The administration of concentrates of vitamin A, both oral and parenteral, was followed by extensive improvement. Cone and rod thresholds were lowered and speed of adaptation was increased. After the vitamin A therapy had been discontinued the improvement of visual adaptation was maintained partially. These findings indicate that abnormal dark adaptation in cases of cirrhosis of the liver is due chiefly to an altered intermediary metabolism of vitamin A.

4. Effects of Drugs and Ingested Substances on Dark Adaptation

I have mentioned in other sections of this presentation the work of Patek and Haig on the effects of the administration of thyroid and of alpha-dinitrophenol upon dark adaptation. The data show a marked increase of speed of adaptation and lowering of visual thresholds after administration of these drugs, both of which increased the basal metabolic rate.

The investigations of Yudkin (1941) indicate that alcohol and benzedrine (amphetamine sulfate) produce a transient improvement of dark adaptation without any corresponding rise of vitamin A in the blood. It is possible in some instances for the resulting improvement to be greater than that produced by vitamin A. Alcohol is a vasodilator and therefore stimulates the shift of blood to the peripheral vessels and to the retina. Amphetamine sulfate is a cerebral stimulant and causes increase of blood pressure. Figures 37 and 38 show the effects of the ingestion of 20 ml of alcohol (in the form of 100 ml

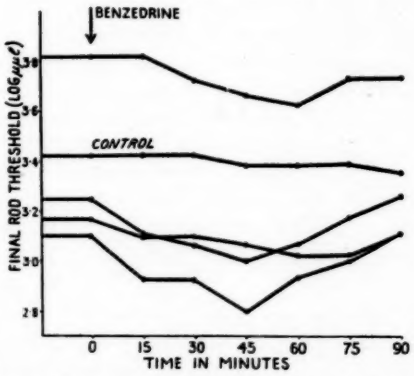


FIG. 38. Final rod thresholds of four subjects to whom were administered 10 mg of benzedrine and of a control subject. (From Yudkin, 1941.)

of sherry) and of a dose of 5 to 10 mg of benzedrine, respectively, on an empty stomach and about thirty minutes before the tests were made. No significant change of the vitamin level of the blood was found. To emphasize the point that the improvement of dark adaptation does not depend on a concomitant rise of the vitamin A of the blood, two subjects were given 120,000 units of vitamin A and tested at intervals

thereafter. There was no definite change in any part of the curves of adaptation although the vitamin A level of the blood rose to more than 500 units for each 100 ml of serum. These same two subjects showed a significant effect from the ingestion of alcohol. Seven of the nine subjects tested by Yudkin after the ingestion of benzedrine showed an improvement of 0.2 to 0.3 log unit in the final rod threshold. The two subjects who did not show any improvement with vitamin A but responded favorably to alcohol also responded to benzedrine. The effect of benzedrine apparently decreased or wore off after an hour and a half and at the time the mental effect was the greatest there was no change of the dark adaptation when compared with the values obtained before the ingestion of the drug.

The mechanism of the actions of alcohol and benzedrine on dark adaptation is none too clear or certain. The effects on dark adaptation are not related necessarily to the amount of subjective effects and, in the case of benzedrine, the general effects are maximal after the influence on dark adaptation has worn off. These effects are not associated with changes of the vitamin A level of the blood; in fact, they may produce an improvement beyond that possible with vitamin A and threshold values may be encountered which are considerably lower (increased retinal sensitivity) than those found in some of the keenest night-seers. However, it is to be stated that alcohol is a readily available and useful source of energy and causes an increase of metabolism. Since increased metabolic rates produce, in general, better levels of dark adaptation, the explanation I have just offered would seem acceptable or at least plausible. Benzedrine acts on the central nervous system and stimulates it into activity. From many clinical observations I am aware that fatigue (indefinite and indefinable as that term may be) is present frequently and that such a drug as benzedrine acts as a "lifter" and permits of keener, more accurate, and more rapid judgments. All of these tendencies are favorable to improvement of dark adaptation.

It appears to me that excellent proof of the hypothesis that it is the ingestion of food (whether it be energy in the form of alcohol, sugar, or other easily assimilated forms such as glucose and egg solution) rather than the effects of the administration of vitamin A on the level of vitamin A in the blood that improves dark adaptation is furnished by some of the results of Yudkin's tests. To quote Yudkin:

> Single doses of about 100,000 I. U. of vitamin A alcohol in gelatin capsules containing 25,000 I. U. each were given on an empty stomach. The effect on the dark adaptation curve was followed during the next 24–36 hours, or longer if the effect was prolonged, *food being taken after 3 hours*. [Italics are mine. C. S.]. . . . The results varied in different subjects. Most commonly

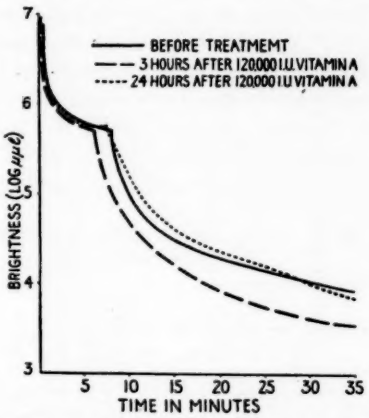


FIG. 39. Dark adaptation curves before and subsequent to the administration of vitamin A to a subject suffering from A avitaminosis. (From Yudkin, 1941.)

> there was an improvement in dark adaptation which began in about 2 hours and *was maximal in about 8 to 10 hours*. [Italics are mine. C. S.] After 24 hours the curve had returned to the original state. This transitory improvement was sometimes sufficient to bring the curve of a severely (?) abnormal subject to within normal limits. [Figure 39 is Fig. 1 from Yudkin's article.] The final rod threshold, the final cone threshold, and the rod-cone time were usually all affected.

I have found on various occasions results almost identical with those of Yudkin in which food, and hence increased metabolic rate, have been responsible for the improvement of dark adaptation, since there was no inclusion of vitamin A in the diet. While the conditions of the experiments are not comparable in all particulars (such as the intensity of the preadapting light), the reader is referred to Fig. 25 of the present paper, in which the dark adaptation of the rods is improved after the ingestion of food, subsequent to an eighteen- to twenty-hour fast, to the same (or a better) level when compared

with the results given by Yudkin and attributed to vitamin A without any consideration of the fact that he fed his subjects during the course of the tests.

That it is the effect of food through increased metabolism rather than vitamin A which is responsible for many of the results reported in the literature and attributed to the vitamin is supported by the investigations of McFarland and Forbes (1940). Figure 40, to which I shall return for further consideration in connection with the effects of oxygen deprivation and hypoglycemia on dark adaptation, shows that when the blood sugar was lowered by insulin the thresholds increased and when the subjects ingested dextrose normal sensitivity returned. It is probable that a deficiency of sugar interferes with oxidative processes which are essential to

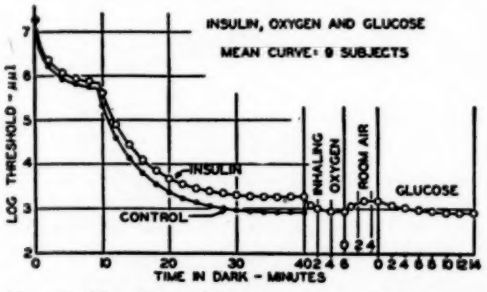


FIG. 40. The effects of insulin, oxygen, and glucose on light intensity. When the concentration of sugar in the blood was lowered by insulin the thresholds increased. (From McFarland and Forbes, 1940.)

neural functioning; hence, in fasting subjects it is possible that the decreased light sensitivity is due to a combination of decreased metabolic rate and an interference with normal neural functioning. If a variation of the content of blood sugar of 20 to 40 mg per 100 cc of blood, which is the usual difference between the basal and the non-basal state, can double the threshold intensity, it would appear that such variables should be controlled in experiments, particularly in tests which may be interpreted incorrectly, or at least questionably, because of failure to control all the known factors and variables.

III. AEROMEDICAL PROBLEMS AND CONSIDERATIONS

It is probable that there is a wealth of accumulated information and data concerning the visual adaptation of aviators under conditions of high altitude and of low temperature which will be made available, in whole or in part, after the days of peace are vouchsafed once again to the peoples of the world. There are several presentations and discussions of an aeromedical character which cannot be committed to paper at the present time by those who are engaged in some of these projects or are acquainted with the contents of confidential communications. However, prior to 1942 there were published reports of researches on the effects of changes of the concentrations of oxygen and of glucose on dark adaptation as well as some scattered investigations on the effects of variations of the content of carbon dioxide of the inspired air and of the rate of respiration on visual discrimination and thresholds.

1. Effects of Anoxia

During the war of 1914–1918, Wilmer and Berens (1918) made the first systematic studies on the effects of lack of oxygen on the aptitude of pilots for flying at high altitude. Standard tests were given to a large number of normal and of defective subjects in a low pressure chamber as well as oxygen deprivation with a rebreathing apparatus. Reports of these findings are contained in a series of papers in the *Air Service Medical Manual*. A large part of the more recent literature has appeared in foreign periodicals but, with few exceptions, the work has not been controlled carefully. In general, however, the investigations showed significant effects of deprivation of oxygen on many visual functions.

The retina is related closely to the brain: embryologically, morphologically, and physiologically. In fact it may be stated that the brain is exposed to the outer world in two places—the retinas of the two eyes. Krause (1934) wrote: "The metabolism of the retina is similar to that of the brain, and not of other tissues. This is to be expected since the retina is anatomically a part of the brain. Weinstein (1932) reported that in the retina and brain the oxygen consumption and carbon dioxide production aerobically and anaerobically are similar." McFarland, Evans, and Halperin (1941) stated:

> Nervous tissue has been shown to be particularly sensitive to a deficit of oxygen. It is consequently not

surprising that investigation of certain functions that involve the retina has revealed that these also manifest extensive changes on exposure of the subject to low oxygen tension. Certain visual characteristics, such as sensitivity to light, are affected more easily than others, such as visual acuity or size of the visual fields. It has not been possible to determine to what extent the effects of anoxia on vision are due to alterations in the central nervous system and to what degree the organ itself is affected. There is an increasing amount of evidence . . . which seems to indicate that the neural components of the retina and of the brain are more extensively involved than was originally supposed, even in such characteristics as sensitivity to light and visual acuity.

It is known in a general way that moderate degrees of lack of oxygen cause a general darkening and narrowing of the visual field and then a blurring of form or a decrease of visual acuity. With increased severity of anoxia there is frequently an intermittent cessation of all visual sensations just previous to loss of consciousness. A general dimming or darkening of the visual field often is reported by pilots while flying at considerable heights (17,000 feet [5200 meters] and above) without oxygen. In laboratory experiments with the Douglas bag it is found that the subject is aware of the dimming of lights on being subjected suddenly to partial pressures of oxygen simulating 12,000 to 14,000 feet (3700 to 4300 meters). There is, on the other hand, an apparent marked increase of the brightness of the lights when the subject is carried back very rapidly to ground conditions. Early investigations indicated definitely that measurements of the thresholds of light sensitivity offered one of the most delicate methods for obtaining information regarding the initial as well as the advanced effects of anoxia. In the experiments with an optical wedge Wilmer and Berens (1918) found that during the rebreathing test the light sensitivity showed an improvement in 25.9 percent of their subjects, no change in 44.5 percent, and a decline in 29.6 percent. During trans-Pacific flights at 11,000 to 12,000 feet (approximately 3500 meters) McFarland and Edwards (1937) observed a decrease of light sensitivity in most of the subjects.

Among the investigations carried on outside of this country are the reports of Tanaka and Sekiguchi (1935), Fischer and Jongbloed (1935), Bunge (1936), Vishnevskiy and Tsyrlin (1935–36), and Clamann (1938). Using a low pressure chamber, Fischer and Jongbloed concluded that there was a slight "delay" of dark adaptation at 3000 meters (about 10,000 feet) and a marked delay at 6000 meters. They interpreted this change as the physiological expression of interference with the regeneration of the photosensitive substance of the retina—an interpretation which is quite likely incorrect, as will be shown in succeeding paragraphs. Bunge administered low oxygen mixtures (8 to 11 percent of oxygen) and found a decrease of retinal sensitivity, with a restoration to normal levels through the administration of oxygen. Since such changes took place in dark-adapted subjects, destruction of visual pigments or a delay in their regenera-

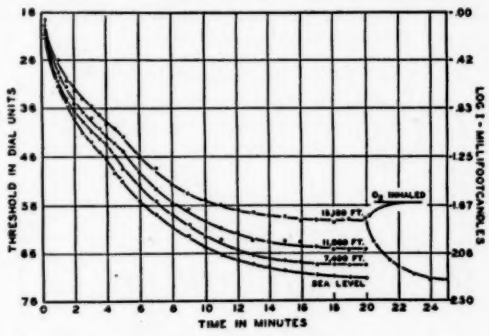


FIG. 41. Alterations of dark adaptation under reduced oxygen tensions. As the oxygen tension is diminished the entire curve is displaced upward, indicating impaired sensitivity. The inhalation of oxygen at 15,100 feet (4600 meters) produced a rapid return to normal levels. (From McFarland and Evans, 1939.)

tion could not have been involved in all probability. Furthermore the return to normal retinal sensitivity under the administration of oxygen was too rapid to be accounted for by regeneration of rhodopsin.

An extensive study of the effects of anoxia on dark adaptation was made by McFarland and Evans (1939) and more recently by McFarland and Forbes (1940–41) and by Brown, Wilson, and myself (1942–43). Twenty subjects were tested by McFarland and Evans at altitudes varying from 7400 to 15,100 feet (about 2300 to 4600 meters) simulated in a low oxygen chamber. Figure 41 shows the composite curve of dark adaptation at each altitude as well as the effects of administration of oxygen at the end of a twenty-minute period of test at an altitude of

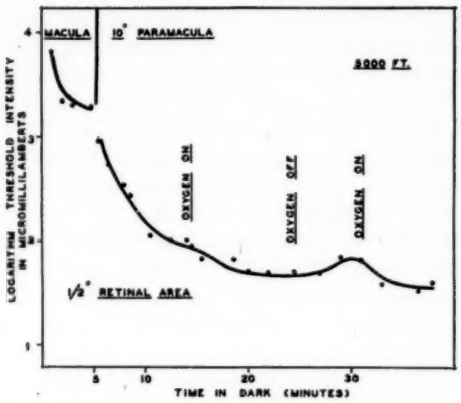


FIG. 42. Dark adaptation thresholds at an altitude of 5000 feet (1500 meters) above ground (6000 feet [1800 meters] above sea level) with and without the use of oxygen. (From Sheard, Brown, and Wilson.)

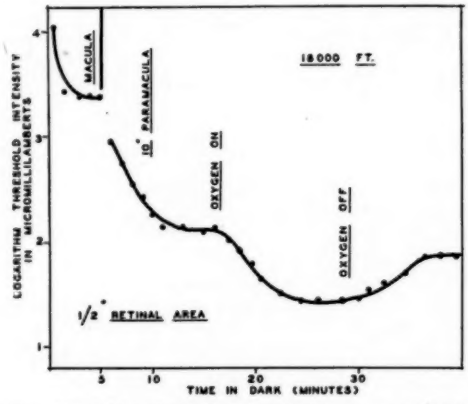


FIG. 43. Dark adaptation thresholds at an equivalent altitude of 18,000 feet (5500 meters), showing the marked advantage of oxygen as compared with air. A threefold to fourfold improvement of levels is demonstrated when oxygen is used. (From Sheard, Brown, and Wilson.)

15,000 feet (4600 meters). At 7400 feet (2300 meters) the mean rise of threshold was 0.1 log unit. At 15,000 feet (4600 meters), the thresholds rose an average of 0.4 log unit: In other words, a light intensity that was two and a half times the normal amount was needed in order to be seen. The shape of the curves is not altered; hence the rate of adaptation is not changed. Brown, Wilson, and I proceeded in a similar manner to substantiate the results of the work of McFarland and Evans as well as of McFarland and Forbes. Using a somewhat different procedure, as is shown in Figs. 42 and 43, we were able to show also that, for some subjects, a difference of sensitivity with and without oxygen could be detected readily and very frequently when the subject was at 5000 feet (1500 meters). Figure 44 is taken from the work of McFarland and Forbes and contains the mean curves of dark adaptation for six subjects at ground level and simulated altitudes ranging from 11,500 to 18,000 feet (3500 to 5500 meters). Table V contains data obtained on the same subject on different days under the same general conditions.

These results emphasize (1) the effects of lack of oxygen on both the cone and rod thresholds of dark adaptation after sufficient lapse of time (twenty-five minutes) to ensure photochemical and pigmentary equilibrium, (2) the degree of reproducibility of data on different days, (3) the beneficial effects of oxygen on the dark adaptation thresholds at ground levels of certain subjects and under untoward environmental circumstances or fatigue, and (4) the general stabilizing action of oxygen. Furthermore, the character of the tests may be varied so that, while each subject is dark-adapted, the concentrations of oxygen may be decreased gradually. Without question regeneration of visual purple will be complete during forty minutes of dark adaptation. Under such circumstances, the thresholds of the subjects tested will continue to rise somewhat in proportion to the degree of anoxia. Inhalation of oxygen rapidly counteracts the effects of a low degree of anoxia, since the thresholds, in general, return to the original control level in two to four minutes. These various curves and data show that anoxia produces a change of

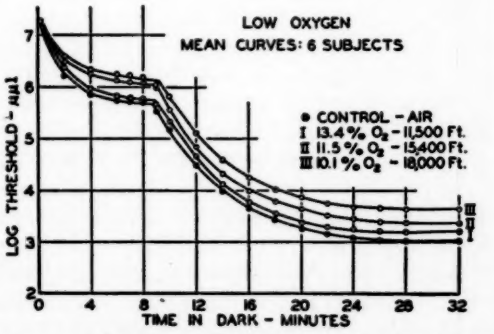

LOW OXYGEN
MEAN CURVES: 6 SUBJECTS
● CONTROL - AIR
I 13.4% O2 - 11,500 Ft.
II 11.5% O2 - 15,400 Ft.
III 10.1% O2 - 18,000 Ft.
LOG THRESHOLD - mμl
TIME IN DARK - MINUTES

FIG. 44. Mean dark adaptation curves showing that, if the oxygen tension was low, the thresholds were high. (From McFarland and Forbes, 1940.)

the cone threshold comparable to, if not equal to, the change of the rod threshold.

McDonald and Adler (1939) investigated the effects of anoxemia on the dark adaptation of the normal and of the vitamin A deficient subject and showed that the normal character of the dark adaptation was altered by a change of two metabolic requirements. The changes produced by anoxia and the deficiency of vitamin A were dissimilar. Whereas anoxia produced equal changes of the rod and cone thresholds, deprivation of vitamin A caused an unequal rise of the rod and cone levels, the rod threshold showing the greater variation. Since it is known that vitamin A is concerned with the photochemical basis of the visual response, it is probable that anoxia acts on the neural mechanism.

2. Effects of Metabolic Changes

I have drawn attention briefly in some of the foregoing paragraphs to the influence of the ingestion of food of a high carbohydrate character and the administration of such substances as thyroid and alpha-dinitrophenol on dark adaptation. Also I have noted the fact that oxygen deprivation and hypoglycemia operate to produce a reduction of retinal sensitivity.

Recently (1940–41) McFarland and Forbes made some observations, under well-controlled conditions, of the effects of oxygen and glucose on the curve of dark adaptation. Typical results are shown in Figs. 45 and 46. The blood sugar averaged 65 mg per 100 cc during the tests with low oxygen. Following the ingestion of glucose (1 g glucose per kilogram body weight in 200 cc of water) the value of the blood sugar rose to an average of 128 mg per 100 cc. In observations in which the blood sugar was raised by ingestion of glucose while the subject was breathing normal air, no significant changes of thresholds occurred except when the blood sugar was falling rapidly toward the end of the glucose tolerance tests. However, when glucose was taken at the end of a test under reduction of oxygen and while the subject remained dark-adapted, the effects of the anoxia were counteracted quite largely in six to ten minutes.

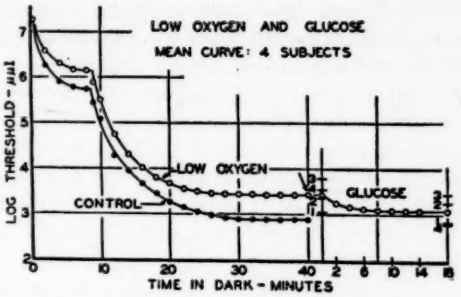


FIG. 45. The effects of anoxia and of glucose on the average dark adaptation curve for four subjects. The solid circles (control curve) are based on measurements obtained in normal air, and the open circles in concentrations of oxygen averaging 10.4 percent. The extent to which glucose counteracted the effects of anoxia on each subject is shown in the difference between the numbered lines at the end of the graph. Each number represents an individual subject. (From McFarland and Forbes, 1940.)

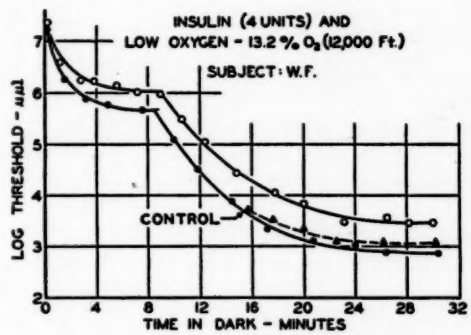


FIG. 46. The combined effects of insulin and low oxygen on light sensitivity. The control curve contains measurements in normal air; the open circles indicate data obtained in reduced oxygen and after the injection of 4 units of insulin. These effects were largely counteracted by the ingestion of glucose (indicated readings are triangles), the oxygen tension remaining at its previous level, 13.2 percent. (From McFarland and Forbes, 1940.)

TABLE V. Cone and rod dark adaptation thresholds at various altitudes with air and with oxygen.* (From Sheard, 1942.)

	Macula		10° Paramacula (1° stimulus area)	
	Air	Oxygen	Air	Oxygen
Ground (1000 feet)	3.62	3.61	1.57	1.51
Ground (1000 feet)	3.62	3.58	1.57	1.50
5000 feet	3.64	3.56	1.66	1.45
Ground (1000 feet)	3.43	3.16	1.67	1.48
10,000 feet	3.45	3.24	1.92	1.37
Ground (1000 feet)	3.36	3.24	1.55	1.37
15,000 feet	3.49	3.18	2.00	1.38

* The values in the table are the logarithms of the dark adaptation threshold intensities in micromillilamberts.

The effects of basal and non-basal states on the tests of dark adaptation are illustrated further by the data in Table VI concerning the thresholds of light sensitivity based on the means of ten subjects in the basal and non-basal state while they were breathing normal air. It is well known that the blood sugar may be as low as 70 to 80 mg per 100 cc in the morning and that, after a normal breakfast, it may rise to 100 to 120 mg per 100 cc. The data of Table VI show that the final threshold in the rod portion of the mean curve was 0.3 log unit higher in the basal than in the non-basal state. As commented elsewhere, it is possible that the intra-individual variation during the day or from day to day may be accounted for on the basis of the metabolic state.

The experimental results obtained by several groups of investigators lend support to the hypothesis that the effects of anoxia and hypoglycemia on light sensitivity are exerted on the nervous tissue of the visual mechanism and on the connecting pathways from the retina to the cerebral cortex rather than on the photochemical substances of the receptor organs of the retina.

3. Respiratory Effects

As has been shown, the absolute threshold of vision of a completely dark-adapted person remains constant within narrow limits. The photochemical systems of the receptors are at rest. Under such circumstances various types of physiological stress cause appreciable changes in the thresholds of the dark adaptation which originate at levels central to the photochemical system itself. Hence, in this sense, the threshold of the dark-adapted eye is an indicator of central nervous imbalance. In preceding paragraphs I have outlined the tests which demonstrate that a lowering of the oxygen tension of the inspired air to about 10 percent raises the threshold twofold to fourfold. Hypoglycemia has been reported to induce a similar response. It remains to discuss the possible effects of changes in the rate of breathing and the composition of the inspired gases on the thresholds of the completely dark-adapted eye.

TABLE VI. Thresholds of light sensitivity based upon the means of ten subjects in the basal and non-basal state in normal air. (From McFarland and Forbes, 1940.)

Time (minutes)	Basal threshold (Log I)	Non-basal threshold (Log I)
2	6.33	6.28
4	5.93	5.88
6	5.84	5.84
7	5.75	5.62
8	5.50	5.25
9	5.23	4.85
10	4.91	4.52
12	4.37	3.97
14	3.97	3.55
16	3.68	3.25
18	3.47	3.04
20	3.32	2.94
22	3.30	2.87
24	3.20	2.86
26	3.15	2.84
28	3.15	2.84
30	3.15	2.84

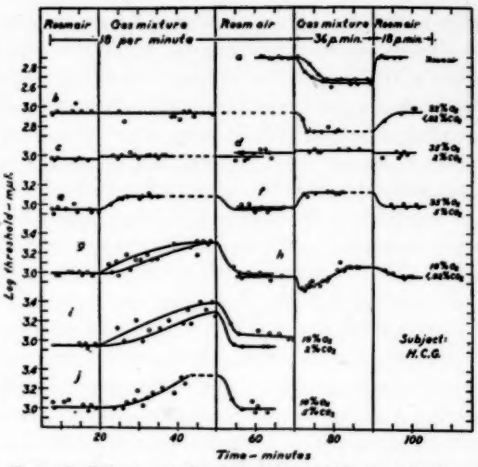


FIG. 47. Effects on the visual threshold of variations of the rate of breathing and of the composition of the inspired gases. Open circles, right eye; solid circles, left eye. (From Wald, Harper, Goodman, and Krieger, 1942.)

Wald, Harper, Goodman, and Krieger (1942) investigated the effects of respiratory acid-base imbalance on the threshold. Some of their findings are shown in Fig. 47. Increasing the rate of breathing room air by 50 to 100 percent caused the threshold to fall to about half its normal value in five to ten minutes (Fig. 47, *a*). In twenty-two experiments with two subjects, this decrease ranged from 0.13 to 0.41 log unit or a decline of about 50 percent. On returning abruptly to the normal rate of breathing it was found that the threshold rose to normal within two to three minutes. [Gellhorn (1936) reported that rapid breathing raises the threshold for brightness discrimination. There is apparently

lack of agreement or else the relation of these two independent investigations is uncertain.] Oxygen tensions of 32 to 36 percent do not affect these results appreciably (Fig. 47, *b*); hence the hypersensitivity induced by rapid breathing is not due to increased oxygenation of the blood. According to Wald and his collaborators, this induced hypersensitivity is due to the alkalosis associated with hyperventilation and can be abolished or reversed by adding carbon dioxide to the inspired gases. The presence of 2 percent of carbon dioxide in mixtures containing 32 to 36 percent of oxygen does not affect the threshold when the mixture is breathed at the normal rate but it abolishes the decrease of threshold otherwise associated with rapid breathing (Fig. 47, *c* and *d*). When the content of carbon dioxide is raised to 5 percent the threshold rises 0.2 to 0.5 log unit at both normal and rapid rates of ventilation (Fig. 47, *e* and *f*). Therefore respiratory alkalosis and acidosis exert opposite effects on the threshold of dark adaptation. Alkalosis depresses and acidosis raises the threshold in about equal amounts, averaging in these experiments about 0.3 log unit or a factor of about two.

Breathing 10 percent oxygen at the normal rate also approximately doubles the threshold. This effect is compensated partially by increased rate of breathing. If 10 percent oxygen is breathed at twice the normal rate, the threshold ordinarily falls at first and then slowly rises to supernormal levels. It is not desirable to discuss here the changes of the blood and tissues which accompany anoxia and changes of respiratory rate. These stresses are a part of a complex pattern of interrelationships. Anoxia generally stimulates rapid breathing owing, in part, to the acidosis which results from incomplete oxidation of metabolic products. In turn, hyperventilation tends to compensate at first for the anoxia. By raising the tension of alveolar oxygen and lowering the tension of alveolar carbon dioxide, it promotes the oxygenation of hemoglobin in the lungs. However, hyperventilation also induces alkalosis, which may intensify the anoxia. This may occur in either of two ways: through the inhibition of the discharge of oxygen from blood to tissues or by causing a period of apnea.

As Wald and his associates pointed out, these various experimental results concerning the effects of anoxia, metabolism, and respiratory rates lend support to the thesis that the visual threshold offers a very practicable quantitative index of physiological imbalance. It is more sensitive and reliable for this purpose than any subjective awareness of distress or sensory change. It has an advantage over certain other types of physiological measurement, for it provides an index of the change in the central nervous system itself—usually the first to fail in physiological stress.

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